ESTACIÓN BIOLÓGICA DE DOÑANA
Dpto. de Biología Aplicada

FACULTAD DE CIENCIAS
Dpto. de Ecología

TESIS DOCTORAL

EFECTOS DE LAS AVES ACUÁTICAS SOBRE LOS MACRÓFITOS Y LOS INVERTEBRADOS EN LAS MARISMAS DE DOÑANA

Memoria presentada por **Héctor Rodríguez Pérez** para optar al grado de Doctor en Ciencias.

Vº Bº del Director:

Fdo. Andy J. Green
Investigador
Estación Biológica de Doñana

***

VºBº del Tutor

Fdo. Ángel Baltanás Gentil
Profesor
Dpto. de Ecología

2006

ÍNDICE:

INTRODUCCIÓN

INTRODUCCIÓN GENERAL:

A pesar de que las aves hayan sido habitualmente un grupo importante de organismos para que el hombre considerara los humedales como zonas valiosas, ya fuera como fuente de alimento, o por el contrario, como motivo para conservarlas (Convenio Ramsar 1971), resulta paradójico que existan pocos trabajos que integren a las aves acuáticas en el funcionamiento de los humedales. Y es que tradicionalmente las aves acuáticas han sido mayoritariamente ignoradas como integrantes de los humedales en las publicaciones de limnología, y no se ha reconocido el papel que desempeñan en los ecosistemas de aguas poco profundas. Esto es especialmente llamativo si se compara con la multitud de publicaciones sobre otro grupo de vertebrados que aparecen en este tipo de publicaciones, como son los peces (Green y Figuerola 2005). Por otro lado, los ornitólogos que dedican sus estudios a las aves acuáticas, tampoco han prestado mucho interés en las interacciones que tienen las aves con otros integrantes del medio, y se han centrado más en estudio sobre la autoecología de determinadas especies, o en tratar de explicar donde se producían grandes concentraciones de aves y porqué seleccionan preferentemente un hábitat frente a otro (fuente de alimento, áreas de cría, etc), sin imbricar en sus estudios aspectos funcionales del hábitat con la actividad de las aves.

Quizás una de las posibles explicaciones de que se haya visto a las aves como organismos ajenos a los sistemas acuáticos, al menos en lo que a su funcionamiento se refiere, sea su capacidad de desplazarse a voluntad lejos de la masa de agua de partida y el que no vivan en el agua sumergidas, por comparación con el resto de los organismos que habitan en el humedal. Y sin embargo los insectos, como los escarabajos o los chinches acuáticos, que completan una parte de su ciclo vital en el agua, en su etapa de imago sí se desplazan fuera del agua volando, o los anuros, que hacen lo propio por tierra. Sin duda en este sentido se puede considerar a todos los grupos anteriormente citados como anfibios, en un sentido amplio, y la única diferencia sería la distancia recorrida. Green y Figuerola (2005) proponen también como posible explicación, que en la Limnología clásica los sistemas se consideran 'cerrados' y al incluir a las aves es necesario considerarlos 'abiertos' y conectados con otros.

En todo caso, esta memoria pretende aportar más datos de los que ya existen, para resaltar la importancia de incluir a las aves como actores importantes en los estudios globales de humedales. Sin duda es necesario, como apuntan Robledano *et al* (1992), mejorar el conocimiento del papel de las aves acuáticas en los ecosistemas en los que habitan, de este modo se podría determinar mejor la 'capacidad de carga' de los humedales que son intensamente ocupados por aves, y valorar como un incremento en las poblaciones de las mismas podría tener consecuencias negativas para el humedal en cuestión, p.ej. al

consumir la mayor parte de la producción del sistema o causar eutrofización. Incluso estos efectos negativos en el humedal podrían extenderse a otras especies de aves amenazadas y muy vinculadas a tipos concretos de humedales, como la cerceta pardilla (*Marmaronetta angustirrostris*), la focha cornuda (*Fulica cristata*), etc. Este aspecto es más relevante aún si se considera la reducción y degradación de las zonas palustres (Jones and Hughes 1993, Green *et al* 2002) y el aumento general de las poblaciones de muchas especies de aves acuáticas (Martí y del Moral 2002).

A la hora de evaluar su importancia para el funcionamiento de los humedales, es importante señalar como características de muchas especies de aves acuáticas, su gran tamaño corporal en relación con muchos de los otros animales con los que comparten hábitat, su alta tasa metabólica como organismos endotermos, y el carácter gregario de algunas especies, que llegan a formar bandos de miles de individuos.

A pesar de lo expuesto anteriormente existen algunos trabajos, cada vez más numerosos, que consideran las funciones de las aves en estos ecosistemas. Dos series de trabajos son fundamentales en este subárea de la ecología acuática. Vareschi y colaboradores publicaron una serie de trabajos sobre la ecología del lago Nakuru en Kenya en los que se integraban los efectos de las aves, así como del resto de organismos, en el funcionamiento general del lago (Vareschi 1978, Vareschi and Vareschi 1984, Vareschi and Jacobs 1984, Vareschi and Jacobs 1985). Y en 1983 Hurlbert y Chang publicaron un trabajo fundamental en el que acuñaron el término 'ornitolimnología', que trataba sobre el control de los flamencos andinos (*Phoenicoparrus andinus*) de los productores primarios bentónicos en un saladar altoandino de Bolivia. La filosofía de este último trabajo es la que inspira la tesis que aquí se presenta.

Aspectos funcionales de las aves acuáticas en ambientes palustres

En uno de los libros de texto de referencia sobre Limnología en su última edición, '*Limnology. Lake and River Ecosystems*' (Wetzel 2001), tan sólo se cita a las aves acuáticas por dos implicaciones en el funcionamiento de los ecosistemas acuáticos; por el carácter herbivoro de muchas especies y por la función de fertilización del sistema con las heces. Sin embargo además de estas dos implicaciones muy importantes, ha quedado demostrado su papel en la dispersión de otros organismos acuáticos, también el control que ejercen sobre las redes tróficas y la modificación de las condiciones fisico-químicas del sedimento, como se expone a continuación. Estos aspectos quedan reflejados en el modelo conceptual mostrado en la figura 1.

Lodge *et al.* (1998) en un trabajo de revisión sobre herbivoría en macrófitos, destacaban el interés que se había dedicado en las publicaciones científicas al impacto de

los insectos sobre los macrófitos, y la poca importancia que se había otorgado a los impactos producidos por aves acuáticas y otros herbívoros con efectos más conspicuos. Y sin embargo hay varios ejemplos que relacionan el declive de poblaciones de aves herbívoras con la disminución en los macrófitos (Hargeby *et al* 1994, Van Donk *et al* 1994), que subrayan la importancia que tiene la presencia de plantas acuáticas para muchas especies de aves. Además, muchos de los trabajos sobre la herbivoría de las aves acuáticas, sugieren que sólo se producen efectos importantes en sistemas de zonas templadas, durante los periodos iniciales de crecimiento de los macrófitos o en otoño cuando la producción es menor, y/o en situaciones en las que se producen concentración de aves debido a movimientos migratorios (Mitchell and Perrow 1998, Marklund et al. 2002).

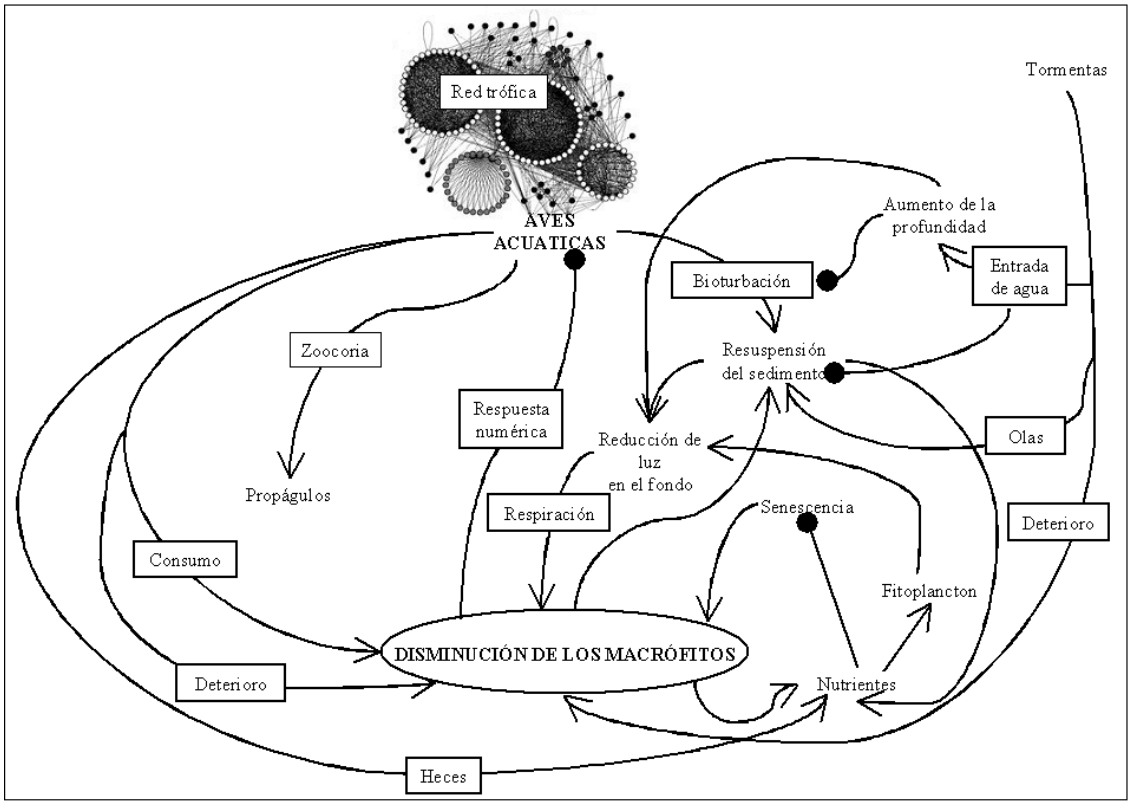


Figura 1.-Modelo conceptual de las funciones de las aves acuáticas en los humedales. Por motivos de claridad del diagrama, las relaciones tróficas de las aves se representan con dibujo de la red trófica; sólo aparece de forma explícita las relaciones con macrófitos, dispersión y parámetros fisico-químicos. Elaborado a partir de la modificación del publicado por (Mitchell and Wass 1996). Las flechas (↑) indican un efecto potenciador, y los símbolos acabados en un círculo (⇡), efectos inhibitorios.

Debido a la importancia que tienen los macrófitos regulando aspectos clave del funcionamiento de los humedales como los flujos de materia y energía, la estabilización del sedimento, la producción primaria y su función de sostén y refugio para muchos otros organismos (ver Jeppensen et al 1998 para una revisión), los cambios en su abundancia

producidos por aves acuáticas tienen consecuencias importantes en todo el sistema. Y existen bastantes trabajos que muestran reducciones significativas de la biomasa de los macrófitos por efecto de las aves, por ejemplo Marklund et al (2002) analizaron una veintena de trabajos en los que no encontraron una relación significativa entre la reducción de biomasa de macrófitos y la abundancia de anátidas y fochas. Pero además de la reducción directa por herbivoría, las aves pueden dañar los macrófitos desenraizándolos, al tirar de los tallos para consumir una parte del mismo o por acción directa del pisoteo (Mitchell y Perrow 1998), pero este tipo de efectos no está bien documentado en la literatura científica. Debido a la importancia de los macrófitos para los humedales, como se ha comentado antes, su eliminación puede acarrear efectos negativos también en otros organismos que dependen de ellos, y bien por depredación directa o indirecta las poblaciones de invertebrados se pueden reducir como demostraron Marklund y Sandsten (2002) y Sherfy y Kirkpatrick (2003) para el caso de cisnes y gansos.

A pesar de que la fertilización por heces o 'guanotroficación' de un humedal aparece en los libros de texto, no resulta tan frecuente encontrar trabajos que estimen su importancia en el balance general de nutrientes. En general, es un proceso que depende de la densidad de aves presentes en el humedal para que suponga un aporte importante de nitrógeno y fósforo al sistema. Así Marion et al (1994) para el lago Grand–Lieu, en Francia, estimaron un aporte anual de la población de aves acuáticas de 0.4 y 0.7% del nitrógeno total y 2.4 y 6.6% del fósforo total en dos periodos de estudio. O Mitchell y Wass (1995) estimaron el aporte de fósforo soluble que hacían los cisnes negros (*Cygnus atratus*) en un lago neocelandés entre 5 y 10 mg/m^3, para una media anual de fósforo total de 340mg/m^3. Las aportaciones al balance total del sistema en estos dos ejemplos no son mayoritarias, pero hay que considerar que ambos ejemplos se establecieron a partir de una población residente de aves. Sin embargo Manny et al (1994) estimaron que el aporte de la concentración de barnaclas canadienses (*Branta canadiensis*) invernantes en el lago Wintergreen, en Estados Unidos, suponía el 27% del nitrógeno total y hasta el 70% del fósforo total. También Portnoy (1990) estableció que dos especies de gaviotas (*Larus argentatus* y *L. marinus*) aportaban 52 kg por año de fósforo total en una laguna costera, y que el 56% de ese aporte correspondía a las concentraciones migratorias en el otoño. Por tanto, este aporte de nutrientes puede ser muy importante para las condiciones tróficas de un humedal, especialmente cuando las aves se agrupan durante la migración, en dormideros (p.ej. Bales *et al*. 1994) o en el caso de aves coloniales como los flamencos.

La función de las aves acuáticas como dispersores de organismos acuáticos fue ya apuntada por Darwin (1859), y observada por otros investigadores de forma anecdótica hasta que Proctor, demostró de forma experimental, su importancia en varios trabajos (Proctor 1959, 1961, 1962, 1964, 1968, Proctor and Malone 1965, 1967). Desde entonces

se han realizado diferentes trabajos demostrando la importancia de la endo y la exozoocoria en los desplazamientos de las aves a largas distancias, y también en ámbitos geográficos más reducidos, y que se recogen en dos revisiones recientes sobre la dispersión por aves acuáticas (Figuerola and Green 2002, Green and Figuerola 2005). Merece la pena destacar cómo este transporte pasivo de semillas o formas de resistencia llevado a cabo por las aves puede ser la forma de dispersión de especies exóticas en los humedales (Green *et al* 2005), o incluso que además de propágulos las aves pueden dispersar por endozoocoria insectos (Green and Sánchez 2005), o cómo la estructura genética de las poblaciones de cladóceros se corresponde con las rutas migratorias de las aves (Figuerola *et al* 2005).

Como postula Steinmetz *et al*. (2003), las aves han sido ignoradas como depredadores superiores en las redes tróficas de sistemas acuáticos, sin embargo, como defienden estos autores, pueden tener un papel fundamental en el control de la red trófica, y por tanto en las comunidades vinculadas a esa red. Además del trabajo ya citado de Hurlbert y Chang (1983), existen más ejemplos publicados de comunidades controladas por aves acuáticas, así Hamilton (2000) demostró el papel como especie clave (*keystone*) del éider común (*Somateria mollisima*) en el ecosistema bentónico de la Bahía de Passamaquoddy, en Canada o los efectos de los patos buceadores en una comunidad controlada por una especie exótica, como el mejillón cebra (*Dreissena polimorpha*), en la región de los Grandes Lagos (Hamilton et al. 1994). O también Wootton (1997) demuestra reducciones importantes en el numero de diferentes invertebrados en una zona intermareal rocosa por acción de ostreros (*Haematopus bachmani*), gaviotas (*Larus glaucescens*) y cuervos (*Corvus caurinus*). Este mismo autor señala que los efectos que las aves pueden producir en la estructura de la comunidad, son desproporcionados respecto a la densidad y biomasa de sus poblaciones, ya que presentan tasas metabólicas más altas que los invertebrados depredadores.

La alteración de origen biológico de las condiciones del sedimento o 'bioturbación', puede cambiar de forma importante el funcionamiento del sistema (Thrush 1999), y además de las modificaciones puramente físicas, estas alteraciones tienen consecuencias en las condiciones químicas, que afectan a los organismos bentónicos y al humedal en su conjunto (Sondergaard et al. 1992) (Lohrer et al. 2004). Como en los casos anteriores, si se considera la llegada masiva de una bandada de aves a un humedal, se encuentran ejemplos significativos de los efectos de las aves como 'bioturbadores'. (Faas et al. 1993) demostraron cómo los bandos de aves limícolas durante la migración, aumentaban la erosionabilidad del sedimento en una zona mareal. Las alteraciones de orden físico se hacen evidentes para el caso del flamenco rosa (*Phoenicopterus ruber*), que en una de sus formas de comportamiento alimenticio, deja unos cráteres evidentes de aproximadamente un metro de diámetro en el fondo de los humedales que ocupa (ver la figura 1 del capítulo

1). Esta modificación de la topografía del sedimento, además de cambios hidrológicos, puede afectar a la vegetación acuática, como sugieren Duarte *et al*. (1990) y Tourenq et al (2001). Además, los efectos físicos de la remoción del sedimento pueden facilitar el acceso de otros depredadores a presas a las que normalmente no acceden, como se ha demostrado para el caso de los efectos de las nutrias marinas (*Enhydra lutris*) en Alaska (Kvitek et al. 1992) y de un especie de raya (*Myliobatis tenuicaudatus*) en Nueva Zelanda (Thrush et al. 1994).

También existen ejemplos directos en la literatura científica de las consecuencias químicas de la 'bioturbación' por aves, y así por ejemplo se demostró que el pisoteo del fondo por flamencos rosas modificó el contenido en oxígeno del sedimento en un zona costera de Namibia (Glassom and Branch 1997b). O de tipo indirecto que demuestran que también el pisoteo de esta misma especie produce la resuspensión de sedimentos, incrementando la concentración de nutrientes en la columna de agua (Comín et al. 1997).

El caso de los flamencos

En la familia Phoenicopteridae se describen tres géneros entre los que se reparten cinco especies, dos especies en el género Phoenicopterus (*P. ruber* y *P. chilensis*) dos especies del género Phoenicoparrus (*P.andinus* y *P. jamesi*) y una en el género Phoeniconaias (*P. minor)*. Las especies de estos dos últimos géneros tienen en el pico estructuras de filtración muy finas, que han evolucionado para una dieta muy especializada en microalgas (Jenkin 1957, Vareschi 1978, Hurlbert and Chang 1983), sin embargo, en el caso de *P. ruber* y *P. chilensis* la dieta es más variada y su sistema de filtración es menos fino (Jenkin 1957). Todas las especies son coloniales, por tanto se pueden dar altas concentraciones en los humedales en los que habitan, y precisamente estos hábitos gregarios y la búsqueda continuada de alimento debido a su gran tamaño, tiene consecuencias en los humedales en los que habitan.

La importancia de los efectos de los flamencos en los humedales quedó señalada hace ya 56 años por Gallet (1950), en la siguiente cita:" *El flamenco altera continuamente el fondo de las lagunas costeras, pisoteando y desenraizando la vegetación, incluyendo a ciertas algas como* Ruppia maritima *cuyos largos, y delicados zarcillos albergan muchos pequeños crustáceos. Éstos constituyen el principal alimento de los patos y las fochas, los cuales, en cuanto su comida es destruida, rápidamente desaparecen de la zona. Esta es una de las diferentes formas con las cuales el flamenco ha modificado radicalmente su hábitat, en detrimento de otras especies. Esto es fuente de satisfacción para aquellos que valoran al flamenco por su exquisita carne, o su vistoso plumaje. Otros verían con buenos*

ojos a los flamencos pereciendo, para preservar (para ellos mismos) a los más humildes patos. ¡Esta es la paradójica situación del asunto!."

Algunos de los ejemplos más destacables de trabajos científicos en el ámbito de la 'ornitolimnología' han incluido alguna especie de flamenco. Y en todos ellos queda patente la importancia de estas aves regulando los flujos de energía los sistemas estudiados. Como es el caso del flamenco enano (*P. minor*) en el lago Nakuru, que según (Vareschi 1978) consumió entre el 50-94 % de la producción primaria del lago, constituyendo entre dos y tres veces la cantidad que el resto de los consumidores primarios consumían, durante el periodo de estudio, comprendido entre los años 1971 a 1973. O el ejemplo, ya comentado, del flamenco andino (*P.andinus*), en Bolivia, que como demostró (Hurlbert and Chang 1983) controlaba las poblaciones de productores primarios y también de consumidores meiobentónicos, y aunque para este caso no existe una estima de energética, se puede adivinar la importancia del efecto en los flujos de energía en la zona de estudio. Los autores apuntaron que el efecto sobre el microbentos no sólo se debía a aspectos tróficos, sino también a la combinación de éstos con los efectos del pisoteo en el sedimento y el aporte de nutrientes con las heces. Otro ejemplo sería los dos trabajos de (Glassom and Branch 1997a, Glassom and Branch 1997b), que estudiaron los efectos del flamenco rosa (*P. ruber*) en dos lagunas costeras de Namibia, y demostraron aumentos de hasta tres veces en las poblaciones de la macrofauna del bentos al excluir a los flamencos. Los resultados obtenidos de los efectos sobre la meiofauna bentónica, fitobentos y las propiedades del sedimento no fueron tan concluyentes, aunque sí apuntaban a un cierto efecto de los flamencos. En los tres ejemplos expuestos, en las zonas de estudio se contabilizaron bandos con miles de flamencos.

La especie presente en Doñana es el flamenco rosa, cuya población invernante ha ido creciendo hasta situarse por encima de los 40.000 individuos en 2003 (Aguilera 2004), además, hay registrados eventos de cría en la marisma, y también durante la época de cría, el 80% de los adultos de la colonia de la laguna de Fuentedepiedra se alimenta en Doñana, especialmente en la finca de Veta la Palma (Aguilar-Amat 2003). Esta colonia está situada en la provincia de Málaga a unos 200 km de distancia, en línea recta, y es una de las dos áreas de cría más importantes del Mediterráneo Occidental, junto con La Camarga.

Según Casas y Ramos (1991) esta especie no fue especialmente abundante en el pasado en esta área, y las medidas de gestión encaminadas a favorecer su cría en Fuentedepiedra (Rendón and Jonson 1996), la desaparición de humedales próximos que antiguamente utilizaban como la laguna de la Lantejuela (Montes y Bernués 1991) y la creación de la granja de piscicultura extensiva de Veta la Palma, han podido contribuir al incremento de la población de flamencos de Doñana. A partir de esta situación se han publicado varios trabajos que sugieren que la existencia de grandes bandos de flamencos

en la zona, pueden disminuir la transparencia del agua y la abundancia de macrófitos en varias zonas de la marisma del Parque Nacional de Doñana, provocando pérdidas en la diversidad de macrófitos e invertebrados y alterando el metabolismo del sistema (Duarte *et al*. 1990, Montes y Bernués 1991), aunque no ha habido una demostración experimental.

Esta Tesis.

La siguiente cita de Hurlbert y Chang (1983), resume la justificación esencial para realizar el trabajo de esta tesis: "(...) *consideramos que nuestros resultados son representativos, en un sentido conservador, del impacto que tienen las aves acuáticas en los ecosistemas acuáticos. Sus actividades de alimentación pueden ser intensas en una gran variedad de sistemas acuáticos, especialmente en pantanos, marismas, lagunas costeras, zonas intermareales y zonas submareales someras, ríos, lagunas y lagos someros de zonas de prados, tundras, desiertos, llanuras costeras y otros paisajes. Cualquier experimento bien diseñado que excluya tanto una única especie abundante o todas las especies de aves acuáticas, de forma colectiva, es probable que demuestre la influencia dominante de las aves en dicho hábitat. Su exclusión producirá cambios en la abundancia y distribución, no sólo, de sus presas, sino también de muchas otras especies. Se producirán grandes cambios en la composición taxonómica, la estructura de tamaños y productividad de la comunidad acuática, e incluso en varias condiciones fisico-químicas del sistema. Dicha influencia es probablemente comparable en magnitud a la producida por los peces, la cual se ha demostrado experimentalmente que es amplia* (…)".

Como objetivo principal de esta tesis se planteó la necesidad de comprobar los efectos que las aves acuáticas tenían sobre otros componentes del humedal (macrófitos, invertebrados y trasparencia de la columna de agua), y cómo estos efectos podían afectar al funcionamiento del sistema en Doñana.

La aproximación metodológica consistió en exclusiones selectivas de todas las aves o específicamente de los flamencos, dada la relevancia de esta especie en la zona (ver más arriba). La exclusión de un organismo o un grupo de organismos, es una aproximación clásica en la metodología ecológica que combinada con otras evidencias, aporta información muy valiosa sobre el efecto que la especie o grupo de especies, tienen sobre las presas o el medio. Sin embargo los resultados no establecen por si mismos, de una forma clara, que sólo los efectos de la depredación de los organismos objeto de estudio, sean los causantes de los hallazgos. Los resultados pueden ser una mezcla de efectos producidos por la depredación directa de las especies excluidas, alteraciones del medio, efectos indirectos que favorecen a ciertas especies, etc. Además existen problemas metodológicos en trabajos de este tipo cuando no se establecen controles de los dispositivos de exclusión o no se

controla la 'pseudoreplicación' en el análisis de los datos (ver Sih *et al.* 1985) para una revisión sobre experimentos de este tipo, Hurlbert 1984).

Todos los experimentos de exclusión que se han hecho durante este trabajo se realizaron en dos áreas diferentes de Doñana; en una zona de marisma transformada en el Parque Natural de Doñana (Veta la Palma) y en la marisma natural del Parque Nacional (Lucios de Marilópez y El Lobo), entre los periodos que abarcan de abril de 2001 hasta septiembre de 2002 para los experimentos en el Parque Natural, y desde febrero a julio de 2004 para los del Parque Nacional.

Para no repetir información sobre la descripción de las áreas de estudio, ya incluida en sus correspondientes capítulos, me centraré más en destacar las diferencias más importantes que existen entre ambas áreas, de cara a los experimentos realizados.

Veta la Palma es una finca de unas 10.000 Ha, propiedad de Pesquerías Isla Mayor SA, situada en el término municipal de la Puebla del Río (Sevilla), e incluida en su mayor parte en el Parque Natural de Doñana y rodeada al este por el río Guadalquivir y al este por el Brazo de la Torre. A principios de la década de los años noventa, aprovechando las infraestructuras de drenaje de esa zona de la marisma y antiguos lucios, se construyeron más de una cincuentena de balsas someras para el cultivo extensivo de especies acuícolas (camaron *Palaemonetes varians*, lubina *Dicentrarchus labrax,* albures o lisas *Mugil cephalus* y anguilas *Anguilla anguilla*). Estas balsas se construyeron sobre antiguos lucios naturales de la marisma de Isla Mayor. Con anterioridad se habían inundado de forma permanente 742 Ha para realizar experiencias de cultivos acuícolas durante varios años (Candel 1990). En total hay una superficie aproximada de 3000 Ha inundadas (ver figura 2 del capítulo 2).

Desde su construcción la zona es muy importante para las aves acuáticas, ya que permanece inundada con agua del río Guadalquivir de forma permanente. Para el caso de algunas especies, como los flamencos, la mayor parte de la población que usa el área de Doñana se encuentra en Veta la Palma (Aguilera 2004).

La zona reúne condiciones óptimas para establecer experimentos como los que hemos realizado, ya que además de la abundancia de aves a lo largo del año debida a las condiciones de inundación permanente, se puede considerar cada una de las balsas como grandes mesocosmos de experimenetación, además existen facilidades de acceso a cada una de las balsas por caminos de servicio. El único problema, desde un punto de vista experimental, son los procesos de secado de las balsas para su mantenimiento y para la pesca, que realizan los operarios de la empresa que explota la granja, y que no siempre se producen con la regularidad anunciada de dos años.

La otra zona de estudio se encuentra situada en la Reserva del Guadiamar y de la que se usaron, para realizar los experimentos, los dos lucios más grandes de esa zona, Marilópez y El Lobo, al norte del Parque Nacional de Doñana (ver figura 1 del capítulo 4).

Por contraposición con Veta la Palma, esta zona tiene un régimen de inundación natural que para un año no seco comenzaría aproximadamente en noviembre y finalizaría (dependiendo de las precipitaciones primaverales) entre junio y julio. Esta zona no es usada tan intensamente por las aves como las balsas de Veta la Palma y alberga una mayor riqueza natural y menor salinidad.

Debido a que la inundación de la marisma está sujeta a las variaciones climáticas naturales y a su impredecibilidad existen dificultades para este tipo de trabajos, además, el acceso al tratarse de una zona 'natural' tampoco es fácil. Esta falta de regularidad en los niveles de inundación dificulta también la comparación de resultados entre años; así por ejemplo en el ciclo hidrológico 2003-2004, cuando se realizó uno de los experimentos de este trabajo, la duración de la inundación se prolongó hasta julio y agosto en algunas zonas, sin embargo la sequía severa del ciclo siguiente hubiera imposibilitado la ejecución del experimento.

La estructura de esta tesis consta de cuatro capítulos que se podrían dividir en dos bloques, el primero que incluiría los **capítulos 1, 2 y 3**, que muestran los resultados de los experimentos realizados en Veta la Palma, y en el segundo estaría el **capítulo 4** que, de alguna forma, se puede entender como una manera de contrastar los resultados encontrados en un sistema 'semi natural' (Veta la Palma) con otro 'natural' (la marisma del Parque Nacional). Los tres primeros capítulos, además del área de estudio, comparten diseño experimental y ejecución, aunque no hay solapamiento completo en las fechas.

En el **capítulo 1** se estudia el efecto que tienen las aves sobre la especie de macrófito (*Ruppia maritima*), que domina la comunidad en Veta la Palma. El estudio comenzó en julio de 2001, y se divide en cuatro periodos consecutivos de tres meses, durante los que se instalaron cercados de exclusión (3x3 metros) para flamencos y para todo tipo de aves. El objetivo era cuantificar los efectos de la herbivoría de fochas y patos, así como comprobar los posibles efectos negativos del pisoteo de los flamencos sobre los macrófitos, pero recogiendo los cambios en la abundancia de macrófitos y aves a lo largo de un ciclo completo.

En el **capítulo 2** se estudia el efecto que tienen las aves sobre los invertebrados del bentos. En este caso el estudio comenzó en abril de 2001, y se utilizaron simultáneamente los mismos cercados que en el **capítulo 1**. El objetivo era cuantificar la disminución en las poblaciones de invertebrados debida a la acción de las aves, por depredación o alteración de las condiciones del sedimento o la reducción de los macrófitos.

En el **capítulo 3** se estudia el efecto que tienen las aves sobre los invertebrados de la columna de agua, desde el zooplancton a macroinvertebrados nectónicos, y epifíticos. La ejecución de los experimentos es la misma que en el capítulo anterior. Para este caso la falta absoluta de información previa, aconsejaba inicialmente marcarse el objetivo de

comprobar si las aves tenían efectos sobre la abundancia de estos organismos y cuantificarlos, si existían.

En el **capítulo 4** se estudia el efecto de los flamencos en la marisma natural del Parque Nacional de Doñana. El estudió comenzó en febrero de 2004 y terminó en julio de ese mismo año, y se instalaron cercados de exclusión de flamencos (4x4 metros). En este caso se trató de comprobar los efectos de los flamencos sobre los macrófitos, la turbidez en la columna de agua asociada a la reducción de la vegetación y a la resuspensión del sedimento, y sobre los macroinvertebrados del bentos, eligiendo como organismo modelo las larvas de mosquitos de la familia Chironomidae.

Los capítulos de esta tesis tienen formato de artículo, y por eso es inevitable el que aparezca información redundante en varios de ellos, especialmente en los apartados de descripción de los métodos y los materiales empleados. No obstante, los datos y resultados de cada capítulo son originales y específicos de cada uno.

Referencias:

Aguilera, E. 2004. Reconstrución de la fluctuaciones poblacionales de las aves de Doñana a lo largo de las tres últimas décadas. Informe técnico. Convenio Organismo Autónomo de Parque Nacionales y CSIC.

Amat, J.A. 2003. Dependencia de humedales andaluces para el exito de la colonia de flamencos de la laguna de Fuentedepiedra. Informe técnico. Convenio Consejería de Medio Ambiente de la Junta de Andalucía y CSIC.

Bales, M., Moss, B., Phillips, G., Irvine, K. & Stansfield, J. 1993. The Changing Ecosystem of a Shallow, Brackish Lake, Hickling Broad, Norfolk, UK. 2. Long-Term Trends in Water Chemistry and Ecology and Their Implications for Restoration of the Lake. *Freshwater Biology*, 29, 141-165.

Casas, J., and B. Ramos. 1991. Bases para la valoración de la posible incidencia del flamenco rosa (Phoenicopterus ruber roseus) en los ecosistemas del Parque Nacional de Doñana.:159-164.

Comín, F. A., J. A. Herrera-Silveira, and M. Martín. 1997. Flamingo footsteps enhance nutrients release from the sediment to the water column. Wetlands International Publications. Limnology and waterfowls. Monitoring, modelling and management. Proceedings of a Symposium on Limnology and Waterfowl. 43:211-227.

Darwin, C. (1859). On the origin of species by means of natural selection. John Murray. London.

Duarte, C. M., C. Montes, S. Agustí, P. Martino, M. Bernués, and J. Kalff. 1990. Biomasa de macrófitos acuáticos en la marisma del Parque Nacional de Doñana (SW España); Importancia y factores ambientales que controlan su distribución. Limnetica 6:1-12.

Faas, R. W., J. Grant, B. Long, D. M. Paterson, G. M. E. Perillo, and M. C. Piccolo. 1993. An ecological cascade effect: Migratory birds affect stability of intertidal sediments. Limnology and Oceanography 38:225-231.

Figuerola, J., and Green, A.J. 2002. Dispersal of aquatic organisms by waterbirds: a review a past research and priorities for future studies. Freshwater Biology 47:483-494.

Figuerola, J., Green, A.J., and Michot, T.C. 2005. Invertebrate eggs can fly: evidence of waterfowl mediated gene-flow in aquatic invertebrates. American Naturalist 165:274-280.

Gallet, E. 1950. The flamingos of the Camargue. Blackwell Oxford.

Glassom, D., and G. M. Branch. 1997a. Impact of predation by greater flamingos *Phoenicopterus ruber* on the macrofauna of two southern African lagoons. Marine Ecology Progress Series 149:1-12.

Glassom, D., and G. M. Branch. 1997b. Impact of predation by greater flamingos *Phoenicopterus ruber* on the meiofauna, microflora, and sediment properties of two southern African lagoons. Marine Ecology Progress Series 150:1-10.

Green, A.J., El Hamzaoui M., El Agbani M.A., and Franchimont J. 2002. The conservation status of Moroccan wetlands with particular reference to waterbirds and to changes since 1978. Biological Conservation 104:71-82.

Green, A.J., Sánchez, M.I., Amat, F., Figuerola, J., Hontoria, F., Ruiz, O., and Hortas, F. 2005. Dispersal of invasive and native brine shrimps *Artemia* (Anostraca) via waterbirds. Limnology and Oceanography 50:737-367.

Green AJ and Figuerola, J. 2005. Recent advances in the study of long-distance dispersal of aquatic invertebrates via birds. Biodiversity and Distribution 11:149-156.

Green, A.J., and Sánchez, M.I. 2005. Passive internal dispersal of insect larvae by migratory birds. Biology Letters 2:55-57.

Hamilton, D. J. 2000. Direct and indirect effects of predation by Common Eiders and abiotic disturbance in an intertidal community. Ecological Monographs 70:21-43.

Hamilton, D. J., C. D. Ankney, and R. C. Bailey. 1994. Predation of Zebra Mussels by Diving Ducks - an Exclosure Study. Ecology 75:521-531.

Hargeby, A., Andersson, G., Bindow, I. , and Johansson, S. 1994. Trophic web structure in a shallow eutrophic lake during a dominance shift from phytoplankton to submerged macrophytes. Hydrobiologia 279/280:83-90.

Hurlbert, S. H., and C. C. Y. Chang. 1983. Ornitholimnology - Effects of Grazing by the Andean Flamingo (Phoenicoparrus-Andinus). Proceedings of the National Academy of Sciences of the United States of America-Biological Sciences 80:4766-4769.

Hurlbert, S.H. 1984. Pseudorreplication and the design of ecological field experiment. Ecological Monographs 54:187-211.

Jenkin, P. M. 1957. The Filter-Feeding and Food of Flamingoes (Phoenicopteri). Philosophical Transactions of the Royal Society of London Series B-Biological Sciences 240:401-493.

Jeppensen, E., M. Sondergaard, M. Sondergaard, and K. E. Christoffersen. 1998. The structuring role of submerged macrophytes in lakes. Springer-Verlag, New York.

Jones T.A., and Hughes J.M.R. 1993. Wetland inventories and wetland loss studies- a European perspective. Pages 146-169 in: Moser M., Prentice R.C., and Van Vessen J (Eds.). Waterfowl and wetland conservation in the 1990's- a Global perpective. Proc. IWRB Symp., St Peterburg Beach, Florida. IWRB Special Publication nº 26, Slimbridge.

Kvitek, R. G., J. S. Oliver, A. R. Degange, and B. S. Anderson. 1992. Changes in Alaskan Soft-Bottom Prey Communities Along a Gradient in Sea Otter Predation. Ecology 73:413-428.

Lodge, D.M., Cromin, G., Van Donk E., and Froelich, A.J. 1998. Impact of herbivory on plant standing crop: comparisons among biomes between vascular and non-vascular plants, and among freshwater herbivore taxa. Pages 149-179 in Jeppesen E., Sondergaard, M., Sondergaard, M., and Christoffersen, K. (Eds.). The structuring role of submerged macrophytes inlakes. Springer.

Lohrer, A. M., S. F. Thrush, and M. M. Gibbs. 2004. Bioturbators enhance ecosystem function through complex biogeochemical interactions. Nature 431:1092-1095.

Manny, B.A., Johnson, W.C., and Wetzel. R.G. 1994. Nutrients additions by waterfowl to lakes and reservoirs: predicting their effects on productivity and water quality. Hydrobiologia 279/280:121-132.

Marion, L., Clergeau, P., Brient, L., and Bertru, G. 1994. The importance of avian contributed nitrogen (N) and phosphorus (P) to lake Grand-Lieu, France. Hydrobiologia 279/280:133-147.

Marklund, O., and H. Sandsten. 2002. Reduction of benthic macroinvertebrates due to waterfowl foraging on submerged vegetation during autumn migration. Aquatic Ecology 36:541-547.

Marklund, O., H. Sandsten, L.-A. Hansson, and I. Blindow. 2002. Effects of waterfowl and fish on submerged vegetation and macroinvertebrates. Freshwater Biology 47:2049-2059.

Martí R., y del Moral, JC. (Eds). 2002. La invernada de aves acuáticas en España. Dirección General de la Conservación e la Naturaleza- SEO/BirdLife. Organismo Autónomo de Parque Nacionales. Ministerio de Medio Ambiente. Madrid.

Mitchell, S.F., and Wass, R.T. 1995. Food consumption and faecal deposition of plant nutrients by black swans (*Cygnus atratus*) in a shallow New Zealand lake. Hydrobiologia 306:189-197.

Mitchell, S. F., and Wass, R. T.. 1996. Grazing by black swans (Cygnus atratus Latham), physical factors, and he growth and loss of aquatic vegetation in a shallow lake. Aquatic Botany 55:205-215.

Mitchell, S.F., and Perrow, M.R. 1998. Interactions between grazing birds and macrophytes. Pages 175-196 in Jeppesen E., Sondergaard, M., Sondergaard, M., and Christoffersen, K. (Eds.). The structuring role of submerged macrophytes inlakes. Springer.

Montes, C., and M. Bernués. 1991. Incidencia del Flamenco Rosa (*Phoenicopterus ruber roseus*) en el funcionamiento de la marisma del Parque Nacional de Doñana (SW España).105-110.

Proctor, V.W. 1959. Dispersal of fresh-water algae by migratory water birds. Science 130:623-624.

Proctor, V.W. 1961. Dispersal of *Riella* spores by waterfowl. Bryologist 64:58-61.

Proctor, V.W. 1962. Viability of *Chara* oospores taken from migratory water birds. Ecology 43:528-529.

Proctor, V.W. 1964. Viability of crustacean eggs recovered from ducks. Ecology 45:656-658.

Proctor, V.W. 1968. Long-distance dispersal of seeds by retention in digestive tract of birds. Sciences 160:321-322.

Proctor, V.W., and Malone, C.R. 1965. Further evidence of the passive dispersal of small aquatic organisms via the intestinal tract of birds. Ecology 46:728-729.

Proctor, V.W., Malone, C.R., and DeVleming, V.L. 1967. Dispersal of aquatic organisms; viability of disseminules recovered from the intestinal tract of captive killdeer. Ecology 48:672-676.

Rendón, M., and Johnson, A.R. 1996. Management of nesting sites for Greater Flamingos. Colonial Waterbirds 19(1):167-183

Robledano F., Montes C., y Ramirez-Díaz L. 1992. Relaciones ambientales y conservación de las comunidades de aves acuáticas en la gestión de los humedales del sudeste español. Universidad de Murcia 99 pp.

Sherfy, M. H., and R. L. Kirkpatrick. 2003. Invertebrate response to snow goose herbivory on moist-soil vegetation. Wetlands 23:236-249.

Sih, A., P. Crowley, M. Mcpeek, J. Petranka, and K. Strohmeier. 1985. Predation, Competition, and Prey Communities - a Review of Field Experiments. Annual Review of Ecology and Systematics 16:269-311.

Sondergaard, M., P. Kristensen, and E. Jeppesen. 1992. Phosphorus release from resuspended sediment in the shallow and wind-exposed Lake Arreso, Denmark. Hydrobiologia 228:91-99.

Steinmetz, J., S. L. Kohler, and D. A. Soluk. 2003. Birds are overlooked top predators in aquatic food webs. Ecology 84:1324-1328.

Thrush, S. E. 1999. Complex role of predators in structuring soft-sediment macrobenthic communities: Implications of changes in spatial scale for experimental studies. Australian Journal of Ecology 24:344-354.

Thrush, S. F., R. D. Pridmore, J. E. Hewitt, and V. J. Cummings. 1994. The Importance of Predators on a Sandflat - Interplay between Seasonal-Changes in Prey Densities and Predator Effects. Marine Ecology-Progress Series 107:211-222.

Tourenq, C., Aulagnier, S., Durieux, L., et al 2001. Identifying rice fields at risk from damage by greater flamingo. Journal of Applied Ecology. 38:170-179.

Vandon Deck, De Deckere E., Klein Breteler J.P.G., Meulemans, J.T. 1994. Herbivory by waterfowl and fish on macrophytes in a biomanipulated lake: effects on long term recovery. Verh. Int. Verein. Limnol. 25:2139-2143.

Vareschi, E. 1978. The ecology of lake Nakuru (Kenya). Abundance and feeding of lesser flamingo. Oecologia 32:11-35.

Vareschi, E., and J. Jacobs. 1984. The Ecology of Lake Nakuru (Kenya) .5. Production and Consumption of Consumer Organisms. Oecologia 61:83-98.

Vareschi, E., and J. Jacobs. 1985. The Ecology of Lake Nakuru .6. Synopsis of Production and Energy-Flow. Oecologia 65:412-424.

Vareschi, E., and A. Vareschi. 1984. The Ecology of Lake Nakuru (Kenya) .4. Biomass and Distribution of Consumer Organisms. Oecologia 61:70-82.

Wetzel, R.G. 2001. Limnology: Lake and River ecosystem. 3rd Edition. Academic Press.

Wootton, J. T. 1997. Estimates and tests of per capita interaction strength: Diet, abundance, and impact of intertidally foraging birds. Ecological Monographs 67:45-64.

CAPÍTULO 1.

Efecto de las aves acuáticas sobre *Ruppia mairitima* / Waterbirds effect on *Ruppia maritima.*

Basado en el artículo: Rodríguez-Pérez, H., and Green A.J. 2006. Waterbird impacts on widgeongrass *Ruppia maritime* in a Mediterranean wetland: comparing bird groups and seasonal effects. Oikos 112:525-534.

Resumen

Hemos estudiado el efecto de las aves acuáticas sobre el macrófito *Ruppia maritima* en 11 balsas de piscicultura en el Parque Natural de Doñana. Dos tipos distintos de cercados de exclusión, de 3x3 metros, nos permitieron excluir selectivamente los flamencos o todo el conjunto de aves acuáticas y comparar los efectos del los tratamientos con sus respectivos controles. Se realizaron cuatro experimentos de tres meses de duración durante diferentes momentos de un ciclo anual con distintas densidades de aves. Los flamencos, los patos y las fochas tuvieron efectos aditivos significativos sobre la presencia de tallos y hojas, o de raíces de *Ruppia* en cualquiera de las épocas del año estudiadas. En los cercados en los que hubo *Ruppia*, la biomasa de las partes verdes (hojas y tallos) fue significativamente superior en los cercados que excluían a todas las aves, que en los controles o las exclusiones de flamencos. La presencia y la biomasa de esta planta anual varía de forma significativa entre estaciones, así como la densidad de sus semillas en el sedimento. Los cambios en la densidad del banco de semillas se compadecieron con el consumo por las aves. No hubo interacciones significativas entre tratamiento y periodo experimental para el caso de la presencia de *Ruppia*, la biomasa de las partes verdes, ni para la abundancia de semillas en el banco. Este trabajo es el primer estudio de exclusión que compara los efectos de las aves acuáticas sobre los macrófitos en diferentes épocas del año, y el primero que compara de forma simultánea los efectos de diferentes grupos de aves. Nuestro resultados refutan las sugerencias previas, que aseguraban que los efectos importante de las aves acuáticas se limitaban a regiones templadas y a los periodos iniciales del crecimiento, o bien a los momentos de grandes concentraciones otoñales de aves migrantes. Los flamencos son importante a la hora de estructurar los humedales someros en el Mediterráneo, y probablemente en muchas otras regiones.

Palabras clave: herbivoría, flamencos, anátidas, banco de semillas, ingeniero ecológico.

Abstract

We studied the effect of waterbirds on the submerged macrophyte *Ruppia maritima* in eleven fish ponds within Doñana Natural Park (SW Spain). Separate exclosure designs allowed us to exclude flamingos or all waterbirds from 3x3 m plots within the ponds and compare them with control plots. Four experiments were conducted for three month periods at different points of the annual cycle with varying bird densities. Flamingos and wildfowl

(ducks and coot) had significant negative additive effects on the presence of aboveground (leafs and shoots) or belowground (roots) parts of *Ruppia* at all times of the year. For plots where *Ruppia* was present, aboveground biomass was significantly higher in all-bird exclosures than in controls or flamingo exclosures. Presence and biomass of this annual plant varied significantly between seasons as did the density of seeds in sediments. Seasonal changes in seedbank densities were consistent with consumption by birds. There were no significant treatment x season interactions for *Ruppia* presence, aboveground biomass or seeds. This is the first exclosure study to compare the effects of waterbirds on submerged macrophytes at different times throughout the annual cycle, and the first to compare simultaneously the effects of different bird groups. Our findings refute previous suggestions that major effects of waterbirds are limited to temperate regions and to periods of early growth or when major concentrations of migratory wildfowl are formed in autumn. Flamingos are important in structuring shallow wetlands in the Mediterranean, and possibly many other regions.

Keywords: Herbivory, flamingo, waterfowl, seed bank, ecological engineer.

INTRODUCTION

Submerged vascular plants have a vital role in non-marine aquatic ecosystems, influencing nutrient dynamics and water chemistry, modulating the structure and dynamics of pelagic and benthic food webs, and increasing the physical habitat diversity (see Jeppesen et al 1998 for review). Changes to the status of submerged macrophytes owing to herbivory or other factors can have major consequences for the whole ecosystem (Scheffer et al. 1993, Van Donk and Otte 1996, Perrow et al. 1997).

Most studies of herbivory in aquatic systems have been made on insects, even though other groups such as crayfish, fish and waterbirds cause bigger reductions in macrophyte biomass (Lodge et al. 1998). Although several studies have shown that waterbirds can have an important effect on the standing crop of submerged macrophytes, they have been restricted to wildfowl (ducks, swans and coots) and largely concentrated in temperate areas of North America, Europe and New Zealand (Lodge et al. 1998, Marklund et al. 2002). It has been suggested that major effects of waterbirds on macrophytes may only be observed in temperate waters and during periods of early growth or in autumn when macrophyte productivity is low and wildfowl form migratory concentrations (Mitchell and Perrow 1998, Perrow et al. 1997). There is a lack of information from non-temperate (e.g. Mediterranean) regions to test this hypothesis. There is also a lack of studies comparing effects of wildfowl in different parts of the annual cycle, and addressing the impacts of other waterbirds such as flamingos.

Greater Flamingos (*Phoenicopterus ruber*) have undergone a dramatic increase in population size in the Mediterranean region since 1970, and are now one of the most abundant waterbirds in terms of biomass (Johnson 1997, Wetlands International 2002). Without direct evidence, previous workers (Gallet 1950, Grillas et al 1993, Duarte 1990) have suggested that flamingos trample and uproot beds of widgeongrass *Ruppia* and other macrophytes while feeding in Mediterranean wetlands. Whilst this has not previously been tested experimentally, exclosure experiments elsewhere have shown that flamingos can have a marked effect on benthic organisms (Hurlbert and Chang 1983, Glassom and Branch 1997a, 1997 b). Greater Flamingos are also reported to cause significant damage in ricefields (Tourenq et al. 2001), and we suggest that Greater Flamingos are ecological engineers (Fig. 1). Here we present an exclosure study comparing the effects of Greater Flamingos and wildfowl on *Ruppia maritima* in Doñana, south-west Spain, during distinct three month periods of the annual cycle.

The strength of effects on macrophytes recorded by exclosure experiments depends partly on the timing and duration of the experiment and how it relates to the macrophyte life cycle (Mitchell and Wass 1996). It also depends on waterbird phenology. As far as we know,

ours is the first exclosure study to compare the effects of waterbirds on submerged macrophytes at different times throughout the annual cycle. It is also the first exclosure study to address simultaneously the impacts of different groups of waterbirds using the same area, or to assess the influence of changes in waterbird density throughout the annual cycle.

Study area

We carried out exclosure experiments in Veta la Palma (36°57'N, 6 °14'W), a private fish-farm divided in 52 regular ponds within Doñana Natural Park. The ponds were constructed in 1992-1993 on top of what was natural marshland in the Guadalquivir estuary. All the ponds are shallow (average 30 cm, maximum depth 50 cm) and flat-bottomed with a total combined surface area of 2997 ha. The fish species cultured are European Seabass (*Dicentrarchus labrax*), Flathead Mullet (*Mugil cephalus*), Gilthead seabream (*Sparus auratus*) and eels (*Anguilla anguilla*) as well as Atlantic ditch shrimp (*Palaemonetes varians*). Each pond is dried out under rotation approximately every two years to extract fish. Ponds are interconnected via canals and permanent flow of water taken from the Guadalquivir estuary maintains dissolved oxygen levels (see Frisch et al. 2005 for more details).

The dominant submerged macrophyte is *R. maritima*, forming extensive beds during spring and early summer. Small patches of *Potamogeton pectinatus* occur in some ponds in years of lower salinity. Most of the shoreline is bare mud and regular dredging to prevent siltation of peripheral canals used to extract fish from the ponds has restricted development of vegetation, which is dominated by *Arthrocnemum macrostachyum* and *Suaeda spp.* with some small patches of *Phragmites australis* and *Scirpus maritimus* in few ponds. Mean monthly air temperature during the study varied from 11.8 ºC in February to 23.5 ºC in July. Mean annual precipitation is 562 mm/yr with a range of 158-1062 mm/yr (Castroviejo 1993). Salinity during our study varied from 7 g/l during winter months of high rainfall to 15 g/l at the end of September, after the dry summer months typical of the Mediterranean region. pH ranged from 9.3 to 10.4.

MATERIALS AND METHODS

Aerial surveys of waterbirds of the whole Veta la Palma estate (2997 ha) were carried out monthly during the study as part of regular surveys of Doñana National Park and its surroundings. All birds are counted, including those on the water and those in flight (most of which are flushed by the plane). Eleven ponds (range 26-114 ha, a total of 697 ha) were selected for the exclosure experiments so as to include ponds that had been flooded for different lengths of time but were not scheduled to be dried out during the experiments. All

exclosures and controls were 3 x 3m in size, and we used two exclosure treatments. One treatment used a 2 cm mesh nylon net tied to a square PVC frame suspended from four iron poles (2 m long, 10 mm diameter) pushed into the pond bottom in each corner. Wires were also attached from the top of the poles to the centre of the net to prevent it from touching the water surface. At the sides of the square frame, 20 cm of net hung vertically down to prevent birds from passing underneath. This design allowed fish to enter but decreased utilisation by ducks, coot and other birds. The netting did not affect water movement. There was no algal growth and shading effects were close to zero.

The second treatment excluded only flamingos by taking advantage of their much greater height. Four iron poles were placed in each corner of the square as before, and a wire tied round them at a height of 70-75 cm from the bottom (i.e. 30-65 cm above the water surface). This height was selected after trials confirming that other birds pass comfortably underneath the wire and freely enter the exclosures. The controls consisted merely of the four iron poles. Eurasian coots *Fulica atra* and ducks were frequently observed feeding inside controls and flamingo exclosures, whereas birds were never observed inside all-bird exclosures. No evidence of waterbird grazing, such as clipped stems, was observed inside the all-bird exclosures.

Each experimental block contained one exclosure of each kind plus a control, separated by 10 m. Treatments were randomly located within each block. There were two experimental blocks in each pond, separated by 25 m. Both blocks were placed in the western part of each pond, aligning all exclosures and controls at an equal distance from the pond edge (approximately 20 m for the first experiment), so as to expose them equally to the dominant southwesterly wind with an equal and relatively low fetch.

Seeds, aboveground and belowground parts were sampled three months after installing the exclosures. When establishing the exclosures, we only sampled the seedbank. This was owing to their small size and the need to limit disturbance to allow us to study effects on the invertebrate community simultaneously (results to be presented elsewhere). The experiment was repeated four times between July 2001 and September 2002, moving the exclosures and controls 10 m in towards the centre of the pond at the beginning of each new experiment. There was no change in depth towards the centre of the ponds. Owing to the lack of a significant fringe of emergent vegetation, there was no windbreak along the pond edge and no gradient in waterbird density from the edge to the centre of ponds that could have influenced waterbird effects as we moved controls and exclosures between experimental periods (see Weisner et al. 1997). There was slight overlap between experimental periods caused by the time required to sample and move exclosures and controls in eleven ponds. This usually took two weeks, but bad weather caused some delays. The first experimental period ran from 13 July to 30 October 2001 (Oct 01 from hereon),

coinciding roughly with the post-breeding period for waterbirds. The second period, from 22 November 2001 to 27 February 2002 (Feb 02 from hereon) corresponded with the wintering period. The third period from 7 March to 6 June 2002 (Jun 02 from hereon) covered the pre-breeding and early breeding periods. The fourth period, from 12 June to 10 September 2002 (Sep 02 from hereon) covered the breeding and early post-breeding period. Each individual exclosure or control was set for 82-92 days in Oct 01, 95-105 days in Feb 02, 88-96 days in Jun 02 and 83-91 days in Sep 02. In a given pond and period, all exclosures and controls were established or sampled on the same day.

One vegetation sample was taken from the north-east corner of each exclosure and control so as to avoid stepping inside and disturbing spots where separate invertebrate samples were taken. The water depth in the sampling points ranged from 10 to 39 cm (mean ± s.e. = 27 ± 8 cm). A PVC pipe section of 40 cm diameter was inserted into the mud and all the water extracted. Then all *Ruppia* stems and leaves (hereafter aboveground parts) were carefully cut at the base with finger nails and removed. Then three cores of mud were taken from within the pipe area to study roots (hereafter belowground parts). Each core sample was of 5.5 cm diameter and 10 cm depth. Aboveground parts were rinsed in the lab, dried at 80Cº for 48h, then weighed. The number of seeds attached to inflorescences in the aboveground parts were also counted. Such seeds were only recorded in the Jun 02 samples. Mud samples were rinsed with tap water in a 0.5 mm sieve to extract belowground parts which were then dried and weighed.

Seed bank effects were also studied, counting the seeds in separate core samples. Three cores of 5.5 cm diameter and 5 cm depth were taken in each exclosure. These samples were taken both when installing the exclosures and three months later. Seed bank data were available for an earlier experimental period, from 3 April 2001 to 11 July 2001 (Jul 01 from hereon) coinciding with the waterbird breeding period. In contrast, there were no data for the Sep 02 period.

The total number of ponds used for analyses varied from nine to 11 between experimental periods owing to unforeseen drainage of some ponds. Those ponds which only had data for some of the periods were included in our analyses, as the methods used (see below) were robust. However, we repeated our analyses using only the nine ponds used throughout the study and found no important differences (results not shown).

Statistical methods

We used Generalized mixed Linear Models (GLMs, McCullagh and Nelder 1989) to analyze the effects of treatment (all-birds exclosure, flamingo exclosure or control) and experimental period on *Ruppia*. Pond and block nested within ponds were included as

random factors, using the GLIMMIX macro (SAS Institute 1996). Treatment and sampling period were included as fixed factors of three and four levels respectively. *Post-hoc* analyses of least-squared means were carried out to identify significant differences among levels of fixed factors. Tests on the effects of each predictor were performed using *F*-statistics (Crawley 1993).

Data on presence/absence of aboveground or belowground parts in our samples were analysed via logistic regression with a binomial error and a logit link function. To analyse the effects of treatment and sampling period on samples where *Ruppia* was present, we also conducted GLMs of the non-zero biomass data (log transformed to overcome heteroscedasticity) using an identity link and normal error distribution. Owing to the high proportion of zeros, there was no suitable transformation that enabled analysis of all the data (i.e. including zeros). With aboveground biomass as the dependent variable, belowground biomass (log transformed) was included as a covariable to test the hypothesis that treatment and period influenced the relative amounts of above and below ground parts.

The change in density of *Ruppia* seeds in sediments (log transformed) between the beginning and end of each experimental period was also analyzed in a GLM with an identity link and normal error distribution. For samples with non-zero aboveground part biomass, the number of seeds found on plants in June 2002 was analyzed in a GLM with a poisson error distribution and log link function. The biomass of aboveground or belowground parts was included as a covariable together with treatment to test the hypothesis that waterbirds influenced the relative abundance of seeds for a given plant biomass.

We initially considered pond age (the time for which each pond had been flooded at the beginning of the first experimental period) as an additional predictor (results not shown). Pond age was considered as a fixed factor of four levels corresponding to four age categories (ponds filled for < 6 months, 7-12 months, 13-18 months and >19 months). The partial effect of pond age was never significant ($P > 0.1$ in all cases).

RESULTS

Waterbird counts

Peak numbers of waterbirds in the study site occurred during the post-breeding period, reaching more than 50,000 in November 2001 (Fig. 5). Numbers were also high in winter, and were lowest in March and April (Fig. 5). Numbers of the largely herbivorous coots and ducks followed a similar pattern, peaking in October-November during the post-breeding period. Numbers of flamingos reached a peak in July-August at the height of their breeding season (Fig. 5). In terms of biomass, flamingos were easily the most important group of birds

in the area (Fig. 6). The mean density of ducks and coot in Veta la Palma was 2.6 ha^{-1}. The mean density of flamingos was 3.9 ha^{-1}(densities were calculated based on the total pond area of 2997 ha). The most abundant wildfowl species recorded (in order of decreasing abundance) were *Fulica atra*, *Anas penelope*, *A. strepera*, *A. platyrhynchos*, *A. acuta*, *A. clypeata*, *Netta rufina* and *Aythya ferina*, although *A. penelope*, *A. clypeata* and *A. acuta* were only present in winter. The *Anas* dabbling ducks accounted for 96% of the total number of ducks counted.

Presence/absence of *Ruppia*

The proportion of samples in which *R. maritima* aboveground parts were recorded varied consistently between treatments, with lowest presence in controls, highest in all-birds exclosures and intermediate values in flamingo exclosures (Fig. 2). Both treatment and period had highly significant effects on presence of aboveground parts in a logistic regression (Table 1). Post-hoc tests revealed significant differences between all treatments (Flamingos *vs*. All-birds, t_{213} =-3.40, p=0.0008; Controls *vs.* All-birds, t_{213}=-5.36, p<0.0001; Controls *vs.* Flamingos, t_{213}=-2.55, p=0.01). Among sampling periods, Jun 02 had a significantly higher presence than the others (Oct 01 *vs.* Jun 02, t_{213}=-3.43 , p=0.0007; Feb 02 *vs.* Jun 02, t_{213}=-3.59, p=0.0004; Jun 02 *vs.* Sep 02, t_{213}=4.12, p<0.0001), whereas there was no significant difference among the other three periods. The interaction between treatment and period was not significant ($F_{6,207}$=0.19, p=0.97).

Similar results were observed for the presence/absence of belowground parts, with highly significant effects of treatment and sampling periods (Table 1). Post-hoc tests revealed significant differences between all three treatments, with flamingo exclosures showing intermediate root presence (Control *vs.* Flamingo, t_{213}=-2.32, p=0.021; Control *vs.* All-birds, t_{213}=-4.98, p<0.0001; Flamingo *vs.* All-bird, t_{213}=-3.07, p=0.002). There were also significant differences between most periods, except for Oct 01 vs Jun 02 and Feb 02 vs. Sep 02 (Oct 01 *vs.* Feb 02, t_{213}=2.97, p=0.003; Oct 01 *vs.* Sep 02, t_{213}=4.22, p<0.0001; Feb 02 *vs.* Jun 02, t_{213}=-2.42, p=0.016; Jun 02 *vs.* Sep 02, t_{213}=3.77, p=0.0002). The interaction between treatment and period was not significant ($F_{6,207}$=0.47, p=0.83).

Biomass of *Ruppia*

Only non-zero values were included in analyses of biomass. For aboveground parts, biomass varied seasonally with highest values recorded in all-birds exclosures (Fig. 3). Treatment and sampling period both had highly significant effects (Table 2). Post-hoc tests revealed significant differences between all-birds exclosures and the other two treatments

(Control *vs.* All-birds, t_{67}=-2.77, p=0.007; Flamingo *vs.* All-birds, t_{67}=-5.33, p<0.0001; Control *vs.* Flamingo, t_{67}=1.15, p=0.25). Among sampling periods, Jun 02 had a significantly higher biomass than the others (Oct 01 *vs.* Jun 02, t_{67}=-6.33, p<0.0001; Feb 02 *vs.* Jun 02, t_{67}=-5.27, p<0.0001; Jun 02 *vs.* Sep 02, t_{67}=5.35, p<0.0001). The interaction between treatment and period was not significant ($F_{6,61}$=1.70, p=0.13).

Similar results were recorded for root biomass with both treatment and period having significant effects (Table 2). Post-hoc tests showed that the only significant difference between treatments was that between the two classes of exclosures (Flamingo *vs.* All-birds, t_{50}=-2.94, p=0.005; Control *vs.* Flamingo, t_{50}=0.98, p=0.33; Control *vs.* All-birds, t_{50}=-1.56, p=0.13). Among sampling periods, Jun 02 again had a significantly higher biomass than the others (Oct 01 *vs.* Jun 02, t_{50}=-4.62, p<0.0001; Feb 02 *vs.* Jun 02, t_{50}= -3.66, p=0.0006; Jun 02 *vs.* Sep 02, t_{50}= 3.06, p=0.0036). The interaction between period and treatment was marginally significant ($F_{6,50}$=2.33, p=0.047). For periods Oct 01 and Feb 02, root biomass was lowest in controls and was much higher in all-birds exclosures than in the other treatments. In contrast, in Jun 02 and Sep 02 root biomass was highest in Controls and lowest in Flamingo exclosures.

When including belowground biomass as a covariable, the effects of period ($F_{3,84}$=13.68, p<0.0001) and treatment ($F_{2,84}$=4.26, p=0.017) on aboveground biomass remained significant. The partial effect of belowground biomass was highly significant ($F_{1,84}$=52.71, p<0.0001). Whilst controlling for belowground biomass, aboveground biomass remained significantly higher in Jun 02 than in other periods, and higher in all-bird exclosures (post-hoc tests for treatments: Flamingo *vs.* All-birds, t_{84}=-0.1208, p=0.007; Control *vs.* Flamingo, t_{84}=0.0213, p=0.7119; Control *vs.* All-birds, t_{84}=-0.099, p=0.08).

Ruppia seeds

There were no differences among treatments in the change in seed densities in sediments from the beginning to the end of each period, but there were highly significant differences between periods (Table 3, Fig. 4). Post-hoc analysis showed that the change in seed numbers for Jul 01 was more positive than for other periods (Jul 01 *vs.* Oct 01, t_{551}=7.03, p<0.0001; Jul 01 *vs.* Feb 02, t_{551}=5.11, p<0.0001; Jul 01 *vs.* Jun 02 t_{551}=3.95, p<0.0001). The change in seed numbers for Jun 02 was also more positive than for Oct 01 (t_{551}=-3.07, p=0.002) and for Feb 02 (t_{551}=-1.97, p=0.049). The interaction between period and treatment was not significant ($F_{6,543}$=0.51, p=0.8).

For Jun 02, we found no evidence of a treatment effect on the number of seeds recorded on plants. Treatment did not have a significant effect on seeds when considered on its own ($F_{2,16}$ =2.34, p=0.12) nor while including aboveground ($F_{2,15}$=0.10, p=0.90) or

belowground biomass as covariables ($F_{2,13}$=2.88, p=0.092). Both aboveground ($F_{1,15}$=22.82, p=0.0002) and belowground ($F_{1,13}$=5.98, p=0.029) biomass had a significant effect on seed numbers. Both aboveground and belowground biomass were highly correlated with the number of seeds on plants (n=15, aboveground, r=0.80, p<0.01, $\log_{10}$ seeds per sample = -50.753 +3.218 $\log_{10}$ aboveground biomass (g per sample)); belowground, r=0.89, p<0.01, $\log_{10}$ seeds = -19.546 +92.210 $\log_{10}$belowground biomass (g per sample)).

DISCUSSION

Contrary to previous suggestions (Mitchell and Perrow 1998, Perrow et al. 1997), our study shows that major effects of waterbirds on submerged macrophytes are neither restricted to temperate waters nor to periods of early growth or in autumn when macrophyte productivity is low and wildfowl form migratory concentrations. Previous studies in Europe have concentrated on coot and dabbling ducks, for which the strongest impacts have been recorded during autumn and winter congregations that coincide with the end of the plant growing season (Lodge et al. 1998), or during the initial phase of reestablishment of macrophytes following biomanipulation (Lauridsen et al. 1993, 2003). Waterbirds have previously been shown to inhibit growth of *R. maritima* planted experimentally in our study area (Figuerola and Green 2004).

Despite major seasonal variation in the abundance of *R. maritima* and of waterbirds (especially wildfowl), we found no evidence of seasonal changes in the relative effect of excluding flamingos or all waterbirds on *Ruppia* presence or aboveground part biomass. In the absence of information on plant growth rates and feedback effects, we can not translate our results into accurate measures of the proportion of primary productivity or standing crop that was consumed by birds (Mitchell and Wass 1996). Nevertheless they suggest there is no simple relationship between herbivory effects and herbivore density in our study system. Likewise, Marklund et al. (2002) found no significant correlation between waterfowl density and their effects on submerged vegetation in a meta-analysis of previous studies.

Since the biomass of submerged vegetation was relatively low in our study area and particularly low in autumn and winter, the strong effects of waterbirds at all times provides no evidence for thresholds below which there is no herbivory because birds move on to search elsewhere. Verhoeven (1980) reported a threshold of 8.8 g DW/m^2 (equivalent to 0.94 on the y axis of Fig. 3) for coots feeding on *Ruppia*, a value much higher than the biomass we recorded in three of our four study periods (Fig. 3). Similar values were cited by Mitchell and Perrow (1998) for swans feeding on *Potamogeton* tubers or filamentous algae. The position of thresholds is likely to depend on the abundance of food in alternative patches, and the general low abundance of submerged vegetation in our study system may be one reason

why any thresholds were much lower (see also Marklund et al. 2002). The ability of omnivorous ducks and coot to switch to feeding on invertebrates and seeds is likely to enable them to persist in Doñana despite the lack of submerged vegetation. During winter, the mild temperatures and lower energy demands for birds at Mediterranean wetlands may lead them to stay on depleted feeding grounds and persist partly on fat reserves rather than risking a migration to some other site.

Based on overall means for the whole study (including zero data), we found that *R. maritima* aboveground part biomass in controls was only 22% of that when all birds were excluded and only 71% of that when flamingos were excluded. This figure for all birds is much higher than that recorded in most previous studies of avian herbivory, even though our densities of waterbirds were much lower than most (Lodge et al 1998, Marklund et al. 2002). Such strong effects of birds may partly be due to the combination of wildfowl feeding on aboveground parts and flamingos uprooting plants owing to physical disturbance caused by their feeding behaviour (Fig. 1). Other stressors in our study area such as high salinity (Murphy et al. 2003) or fish (Cardona et al. 2001) may also diminish the capacity of *R. maritima* to respond to damage caused by waterbirds. Although none of fish species present are strict herbivores, their movements and defecation are likely to favour phytoplankton at the expense of macrophytes.

Abundance of *R. maritima* varies considerably between years in our study site, and was higher in 2001 than in 2002 (Figs. 2-4). These were years of good and average rainfall respectively, when 30,000 ha of natural, temporary marshland close to our study site was also flooded from November to June. In drier years, the densities of birds in Veta la Palma are much higher because the temporary marshes are dry while the ponds remain flooded, although with a higher salinity. In such dry years, the effects of birds on *R. maritima* appear to be even stronger (J. Figuerola and A.J. Green unpubl. data).

Many submerged macrophytes are only weakly anchored in the sediments by their belowground parts and are particularly vulnerable to disturbance (Combroux et al. 2001, Capers 2003). Ours is the first study to demonstrate a deleterious effect of Greater Flamingos on submerged vegetation, but our results are compatible with observations by previous authors (Gallet 1950, Hurlbert & Chang 1983, Montes and Bernués 1991) and the capacity of flamingos for modifying their environment (Fig. 1) owing to their treading action during feeding (Johnson 1997). The true impact of flamingos may have been underestimated by our data since wildfowl are likely to have been attracted to feed in flamingo exclosures where *R. maritima* was relatively more available. Conservation measures have led to a roughly tenfold increase in the numbers of Greater Flamingos in the Mediterranean region since the 1960s (Wetlands International 2002). This has coincided with a major loss of wetland habitat over the same period (Finlayson et al. 1992), and the resulting increase in

density is likely to have increased the role of flamingos in structuring aquatic ecosystems in the Mediterranean region.

As well as by grazing and physical disturbance, wildfowl and particularly flamingos are likely to have a further harmful effect on macrophyte growth owing to the nutrients added via faeces and the turbidity caused while feeding (Wass and Mitchell 1998). When feeding, flamingos disturb the sediment, mobilise transfer of nutrients from sediments into the water column and increase the turbidity for some time after they have left (Comin and Herrera 1997, Glassom and Branch 1997b). This increase in turbidity by sediment resuspension reduces the available light for macrophytes, favouring a shift to a turbid water state dominated by phytoplankton (Søndergaard et al. 1992, Scheffer 1993). Our exclosures were too small to study these effects, since feeding activity in adjacent areas also raised turbidity within exclosures.

R. maritima can be either an annual or a perennial (Valdés et al. 1987, Verhoeven 1979), but is annual in our study area with no underground storage organs. This makes it easier to understand the effects of grazing and other waterbird activities on future biomass than for the many macrophytes with storage organs. Although we could not demonstrate a direct effect of avian exclusion on seed production, the strong effect on aboveground and root biomass coupled with the correlation between biomass and seed production makes it clear that more seeds are produced in the absence of birds. Thus waterbird impacts in one year are likely to influence *Ruppia* growth the following year. However, it is not clear how many of the extra seeds produced in the absence of birds would be lost before entering the seed bank, e.g. via consumption by other organisms. Whilst controlling for belowground biomass, the aboveground biomass remained higher in all-birds exclosures. This is likely to be because ducks and coot graze on green parts when they are available and have relatively less impact on root biomass.

We observed a weakly significant interaction between period and treatment for positive belowground biomass values. This suggests paradoxically that exposure to waterbirds reduced presence of roots in Jun 02 and Sep 02 but increased biomass for those samples where roots were present. This result may have been a Type I error.

We were surprised not to detect any influence of birds on seed densities in sediments, since at our study site *R. maritima* seeds are an important component of the diet of ducks and coots (Figuerola et al. 2002) and probably also flamingos (Johnson 1997). A flamingo found dead at Veta la Palma in January 2004 contained 246 *R. maritima* seeds in its gizzard, together with benthic ostracods. It is possible we were sampling areas that were deeper than those preferred by ducks for feeding on seeds (Guillemain and Fritz 2002). However, it seems more likely that the reduced loss of seeds from sediments within exclosures was not detected because of the extreme variation in seed densities in our samples, or because of

continuous horizontal movements of seeds in sediments associated with wave action, fish activity or bird activity. While consuming seeds, flamingos mix up sediments with their treading action, and are likely to bring seeds into the top 5 cm layer we sampled from deeper down, thus making it harder for us to detect seed loss. During autumn and winter periods, we found a major decrease in seed densities during our experiments which is attributable to loss from predation (Fig. 4).

The effects of waterbirds on submerged vegetation we have recorded have major consequences for the functioning of aquatic ecosystems (see introduction). More studies of waterbird effects in non-temperate wetlands and more studies comparing different periods of the annual cycle are required before we can understand when, where and why avian effects on submerged vegetation are important. Flamingos and other non-herbivorous waterbirds should be included in such studies. Flamingos are likely to be very important in structuring coastal wetlands and shallow lakes in large parts of the world, and management measures designed to increase their populations (e.g. Martos and Johnson 1996) may have major implications for ecosystem functioning.

Acknowledgments.

This work was funded by the Department of Environment (Consejería del Medio Ambiente) of the Andalusian Autonomous Government. Héctor Rodríguez Pérez had a PhD grant from the CSIC-I3P programme funded by the European Union Social Fund. Aerial count data were provided by the Equipo de Seguimiento de Procesos Naturales of Doñana Biological Station. María del Mar Delgado and Esther Macarro assisted with field work and the identification of flamingo stomach contents. We thank Luis Santamaría, Ramon Soriguer and two anonymous referees for comments on an earlier version of this manuscript.

References:

Capers, R.S. 2003. Six years of submerged plant community dynamics in a freshwater tidal wetland. - Freshwater Biol. 48:1640-1651.

Cardona, L., Royo, P. and Torras, X. 2001. Effects of leaping grey mullet *Liza saliens* (Osteichthyes, Mugilidae) in the macrophyte beds of oligohaline Mediterranean coastal lagoons. - Hydrobiologia 462:233-240.

Castroviejo, J. 1993. Mapa del Parque Nacional de Doñana; Memoria. Consejo Superior de Investigaciones Científicas. Madrid.

Combroux, I., Bornette, G., Willby, N.J. and Amoros, C. 2001. Regenerative strategies of aquatic plants in disturbed habitats: the role of propagule bank. - Arch. Hydrobiol. 152:215-235.

Comín, F.A., Herrera-Silveira, J.A. and Martín, M. 1994. Flamingo footsteps enhance nutrient release from the sediment to the water column. - In: Faragó, S. and Kerekes, J.J. (ed.), Limnology and waterfowl. Monitoring, modelling and management. Wetlands International. pp. 211-227

Crawley MJ. 1993. GLIM for ecologist. - Blackwell Science.

Del Hoyo, J., Elliot, A. and Sargatal, J. (eds.). 1992. Handbook of the birds of the world. - Lynx Editions.

Duarte, C.M., Montes, C., Agustí, S., Martino, P., Bernués, M. and Kalff, J. 1990. Biomasa de macrófitos acuáticos en la marisma del Parque Nacional de Doñana (SW España); Importancia y factores ambientales que controlan su distribución. - Limnética 6:1-12.

Figuerola, J., Green, A.J. and Santamaría, L. 2002. Comparative dispersal effectiveness of wigeongrass seeds by waterfowl wintering in south-west Spain: quantitative and qualitative aspect. – J. Ecol. 90(6):989-1001.

Figuerola, J. and Green, A.J. 2004. Effects of seed ingestion and herbivory by waterfowl on seedling establishment: a field experiment with wigeongrass *Ruppia maritima* in Doñana, south-west Spain. - Plant ecol. 173:33-38.

Finlayson, M., Hollis, T. and Davis, T. 1992. Managing Mediterranean wetlands and their birds. Proceedings of an IWRB International Symposium, Grado, Italy. International Waterfowl and Wetlands Research Bureau.

Frisch, D., Rodríguez-Pérez, H. and Green, A. J. 2005. Invasion of Doñana Natural Park, South-west Spain, by an exotic marine copepod. - Aquatic Conserv:Mar. Freshw. Ecosyst. 15: in press.

Gallet, E. 1950. The flamingos of the Camargue. Blackwell.

Guillemain, M. and Fritz, M. 2002. Ecomorphology and coexistence in dabbling ducks: the role of lamellar density and body length in winter. - Oikos 98:547-551.

Glassom, D. and Branch, G.M. 1997a. Impact of predation by greater flamingos *Phoenicopterus ruber* on the macrofauna of two southern African lagoons. - Mar. Ecol. Prog. Ser. 149:1-12.

Glassom, D. and Branch, G.M. 1997b. Impact of predation by greater flamingos *Phoenicopterus ruber* on the meiofauna, microflora, and sediment properties of two southern African lagoons. - Mar. Ecol. Prog. Ser. 150:1-10.

Grillas, P., Garcia-Murillo, P., Geertz-Hansen, O., Marbá, N., Montes, C., Duarte, C.M., Tan Ham, L. and Grossman, A. 1993. Submerged macrophyte seed bank in a Mediterranean temporary marsh: abundance and relationship with established vegetation. - Oecologia 94:1-6.

Hurlbert, S. H. and Chang, C.C.Y. 1983. Ornitholimnology: Effects of grazing by the Andean flamingo (*Phoenicoparrus andinus*). - Proc. Natl. Acad. Sci. 80:4766-4769.

Jeppesen, E., Sondergaard, M., Sondergaard, M. and Christoffersen, K.E. (eds.). 1998. The structuring role of submerged macrophytes in lakes. - Spring-Verlag.

Johnson, A. 1997. *Phoenicopterus ruber* Greater Flamingo. - BWP Update 1**,** 15-23.

Lauridsen, T., Jeppesen, E. and Østergaard, F. 1993. Colonization of submerged macrophytes in shallow fish manipulated Lake Vaeng: impact of sediment composition and waterfowl grazing. _ Aquat. Bot. 46: 1-15.

Lauridsen, T. L., Sandsten, H. and Møller, P. H. 2003. The restoration of a shallow lake by introducing *Potamogeton* spp.: The impact of waterfowl grazing. - Lakes & Reservoirs: Research and Management 8: 177-187.

Lodge, D.M., Cronin, G., Van Donk, E. and Froelich, A.J. .1998. Impact of herbivory on plant standing crop: comparisons among biomes, between vascular and among freshwater herbivory taxa. - In: Jeppensen, E., Sondergaard, M., Sondergaard, M. and Christoffersen, K.E. (eds.). 1998. The structuring role of submerged macrophytes in lakes. - Spring-Verlag, pp 149-174.

Marklund, O., Sandsten, H., Hansson, L.A. and Blindow, I. 2002. Effects of waterfowl and fish on submerged vegetation and macroinvertebrates. - Freshwater Biol. 47:2049-2059.

Martos, M. R. and Johnson, A. R. 1996. Management of nesting sites for Greater Flamingos. - Colonial Waterbirds 19: 167-183.

Mc Cullagh, P. and Nelder, J.A. 1989. Generalized linear models. - Chapman and Hall.

Mitchell, S.F. and Perrow, M.R. 1998. Interaction between grazing birds and macrophytes. - In: Jeppensen, E., Sondergaard, M., Sondergaard, M. and Christoffersen, K.E. (eds.), The structuring role of submerged macrophytes in lakes. - Spring-Verlag, pp. 175-196.

Mitchell, S.F. and Wass, R.T. 1996. Quantifying herbivory: grazing consumption and interaction strenght. - Oikos 76:573-576.

Montes, C. and Bernués, M. 1991. Incidencia del Flamenco Rosa (Phoenicopterus ruber roseus) en el funcionamiento de la marisma del Parque Nacional de Doñana (SW España). - In: Pintos, M.R., Prieto, S., Rendón, M. and Johnson, A. 1991. Reunión técnica sobre la situción y problemática del Flamenco Rosa (*Phoenicopterus ruber*)en el Mediterráneo occidental y África Noroccidental. Junta de Andalucía.

Murphy, L.R., Kinsey, S.T. and Durako, M.J. 2003. Physiological effects of short-term salinity changes on *Ruppia maritima.* - Aquat Bot 75:293-309.

Perrow, M.R., Schutten, J.H., Howes, J.R., Holzer, T., Madgwick, F.J. and Jowitt, A.J.D. 1997. Interactions between coot (*Fulica atra*) and submerged macrophytes: the role of birds in the restoration process. - Hydrobiologia 342/343:241-255.

SAS Institute Inc. 1996. SAS System for Mixed Models.

Scheffer, M., Hosper, S.H., Meijer, M.L., Moss, B. and Jeppesen, E. 1993. Alternative equilibria in shallow lakes. - Trends Ecol. Evol. 8:275-279.

Sondergaard, M., Kristensen, P. and Jeppesen, E. 1992. Phosphorus release from resuspended sediment in the shallow and wind-exposed Lake Arreso, Denmark. - Hydrobiologia 228:91-99.

Tourenq, C., Aulagnier, S., Durieux, L., Lek, S., Mesléard, F., Johnson, A. and Martin, J.L. 2001. Identifying rice fields at risk from damage by greater flamingo. - J. Appl. Ecol. 38:170-179.

Valdés, B., Talavera, S., Fernández-Galiano, E. (eds.) 1987. Flora vascular de Andalucía Occidental. - Ketres Ed.

Van Donk, E. and Otte, A. 1996. Effects of grazing by fish and waterfowl on the biomass and species composition of submerged macrophyes. - Hydrobiologia 340:285-290.

Verhoeven, J.T.A. 1979. The ecology of Ruppia-dominated communities in western Europe. I. Distribution of Ruppia representatives in relation to their autoecology. – Aquat. Bot. 6:197-268.

Verhoeven, J.T.A. 1980. The ecology of Ruppia dominated communities in western Europe. III. Aspects of production, consumption, and decomposition. - Aquat. Bot. 8:209-253.

Wass, R.T. and Mitchell, S.F. 1998. What do herbivory axclusion experiments tell us? An investigation using black swans (*Cygnus atratus*) and filamentous algae in a shallow lake. - In: Jeppensen, E., Sondergaard, M., Sondergaard, M. and Christoffersen, K.E. (eds.), The structuring role of submerged macrophytes in lakes. - Spring-Verlag, pp. 282-289.

Weisner, S. E. B., Strand, J. A. and Sandsten, H. 1997. Mechanisms regulating abundance of submerged vegetation in shallow eutrophic lakes. - Oecologia 109: 592-599.

Wetlands International. 2002. Waterbird population estimates. - Wetlands International..

	Aboveground						Belowground					
	Estimate	**SE**	**df_N**	**df_D**	**F**	***p***	**Estimate**	**SE**	**df_N**	**df_D**	**F**	***p***
Intercept	-0.27	0.58					-1.5	0.53				
Treatment			2	213	14.9	**<0.0001**			2	213	12.96	**<0.0001**
Control	-2.62	0.49					-2.13	0.43				
Flamingo	-1.41	0.41					-1.13	0.37				
Period			3	213	7.19	**0.0001**			3	213	7.70	**<0.0001**
Oct 01	0.57	0.53					2.28	0.54				
Feb 02	0.45	0.54					0.97	0.56				
Jun 02	2.25	0.55					2.05	0.54				

Table 1.- Summary of Generalized Linear Models testing the partial effects of treatment (factor of three levels) and period (factor of four levels) on the presence/absence for aboveground and belowground parts of *R. maritima*. Pond and block were included as random factors using GLIMMIX, with binomial error and a logit link. See methods for more details. All-bird exclosure treatment and period Sep 02 were aliased.

	Aboveground						Belowground					
	Estimate	**SE**	**df_N**	**df_D**	**F**	***p***	**Estimate**	**SE**	**df_N**	**df_D**	**F**	***p***
Intercept	0.08	0.24					-0.92	0.36				
Treatment			2	67	14.96	**<0.0001**			2	50	4.68	**0.013**
Control	-0.64	0.23					0.16	0.56				
Flamingo	-0.92	0.17					-1.05	0.71				
Period			3	67	18.86	**<0.0001**			3	50	10.57	**<0.0001**
Oct 01	0.08	0.25					0.55	0.39				
Feb 02	0.17	0.27					0.001	0.41				
Jun 02	1.33	0.25					0.64	0.38				

Table 2.- Summary of Generalized Linear Models testing the partial effects of treatment (factor of three levels) and period (factor of four levels) on the biomass (g/m^2, log_{10} transformed, non-zero values only) of aboveground and belowground parts of *R. maritima*. Pond and block were included as random factors using GLIMMIX, with normal error and a identity link. See methods for more details. All-bird exclosure treatment and period Sep 02 were aliased.

	Seed bank					
	Estimate	**SE**	**df_N**	**df_D**	**F**	***p***
Intercept	2.16	0.014				
Treatment			2	549	0.07	0.94
Control	-0.004	0.01				
Flamingo	-0.0007	0.01				
Period			3	549	17.35	**<0.0001**
Jul 01	0.06	0.01				
Oct 01	-0.049	0.02				
Feb 02	-0.017	0.02				

Table 3.- Summary of Generalized Linear Models testing the partial effects of treatment (factor of three levels) and period (factor of four levels) on the change in density of *R. maritima* seeds in sediments from the beginning to the end of each period (n/m^2, log_{10} transformed). Pond and block were included as random factors using GLIMMIX, with normal error and a identity link. See methods for more details. All-bird exclosure treatment and period Jun 02 were aliased.

Figure 1. Aerial photograph of craters made by Greater Flamingos feeding in the sediments of ponds in Veta la Palma. The flamingos were flushed immediately before the photo was taken. Author Héctor Garrido/Equipo de Seguimiento de Procesos Naturales.

Figure 2. The percentage presence of *Ruppia maritima* aboveground parts in the three treatments (all-bird exclosures, flamingo exclosures and controls) at the end of each experimental period. Dates refer to the end of the three month experiments.

Figure 3. Aboveground *R. maritima* biomass (g/m^2, log_{10} transformed) at the end of each experimental period, including all data (i.e. not excluding zeros as for Table 2). C control, F flamingo exclosure and A all-bird exclosure. The numbers of seeds recorded in June 2002 refer to the total numbers of seeds counted in inflorescences. Note the change of scale on the *y* axis.

Figure 4. Changes in *R. maritima* seed densities (mean + s.e.) recorded in the top 5 cm of sediments from the beginning to the end of each period. Light bars show seed densities at the beginning of each period and dark bars at the end. All treatments were pooled together.

Figure 5. Aerial counts of waterbirds in Veta la Palma from March 2001 to October 2002. Totals represent all birds counted in the area, including shorebirds, gulls, herons, etc.

Figure 6. Mean biomass of flamingos and wildfowl during experimental periods in Veta la Palma. Calculations were based on the average aerial counts for each species for each period and body mass as cited by del Hoyo et al. (1992). Dates refer to the end of the three month experiments.

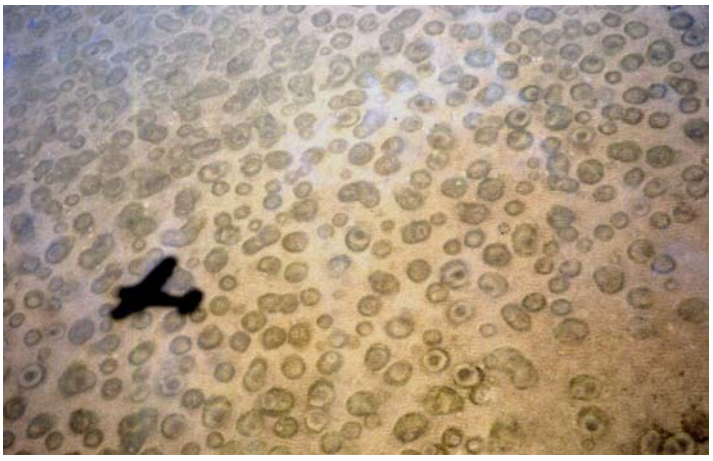


Fig1.-

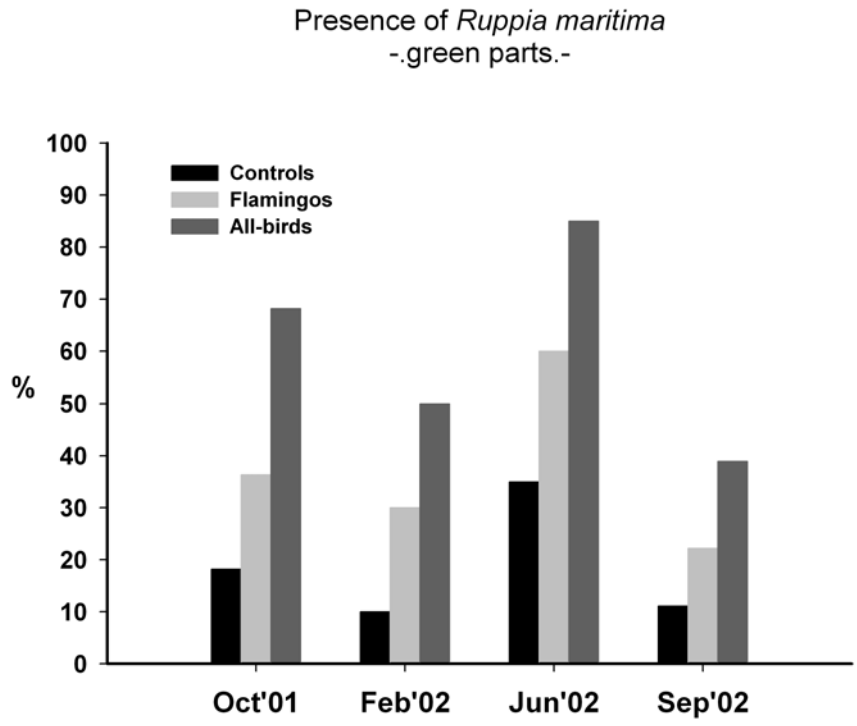

Presence of *Ruppia maritima*
-.green parts.-
Controls
Flamingos
All-birds
%
100
90
80
70
60
50
40
30
20
10
0
Oct'01
Feb'02
Jun'02
Sep'02

Fig 2.-

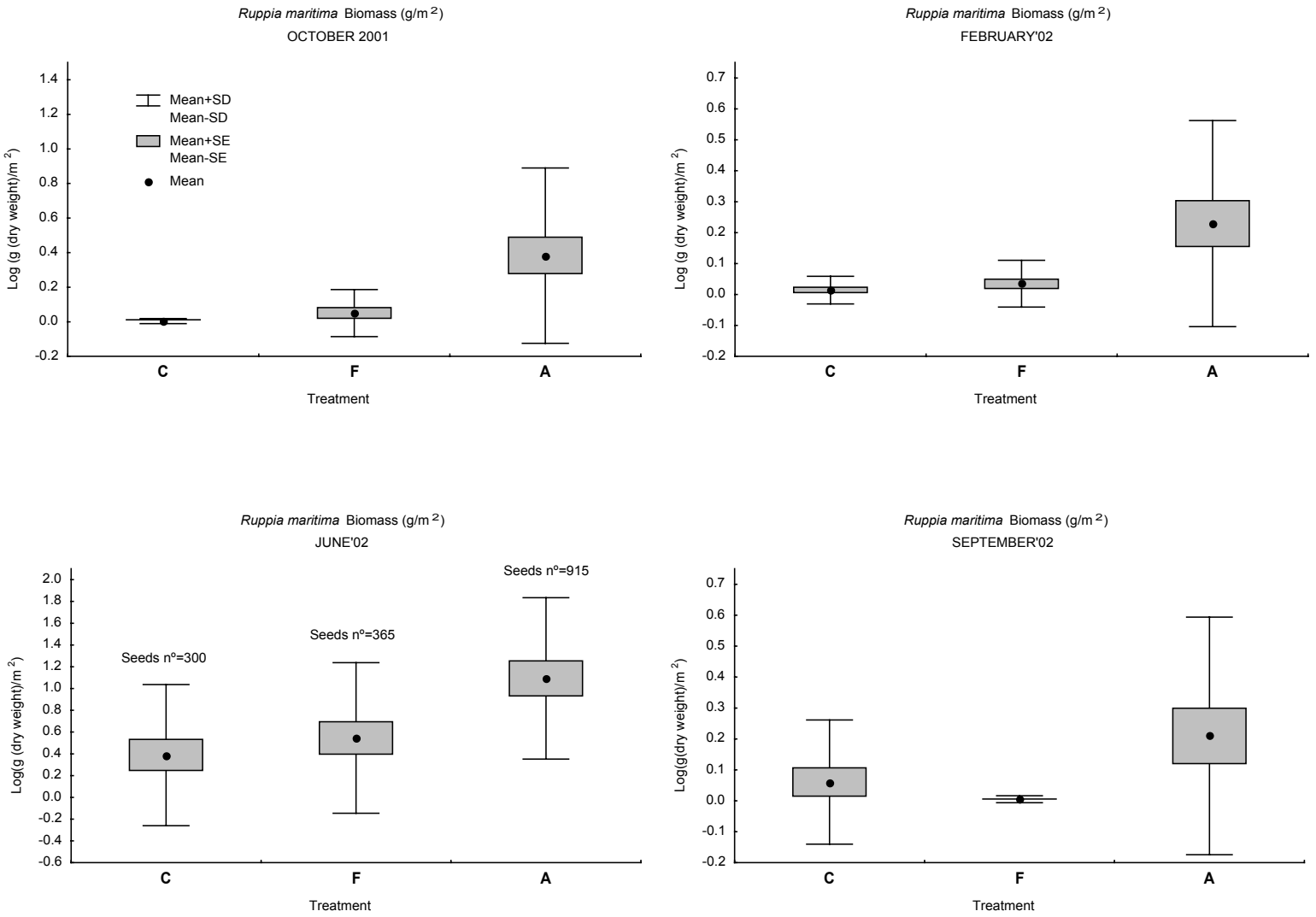

Ruppia maritima Biomass (g/m 2)
OCTOBER 2001
Mean+SD
Mean-SD
Mean+SE
Mean-SE
Mean
Log (g (dry weight)/m 2)
Treatment
C
F
A
Ruppia maritima Biomass (g/m 2)
FEBRUARY'02
Log (g (dry weight)/m 2)
Treatment
Ruppia maritima Biomass (g/m 2)
JUNE'02
Seeds nº=300
Seeds nº=365
Seeds nº=915
Log(g (dry weight)/m 2)
Treatment
Ruppia maritima Biomass (g/m 2)
SEPTEMBER'02
Log(g(dry weight)/m 2)
Treatment

Fig 3.-

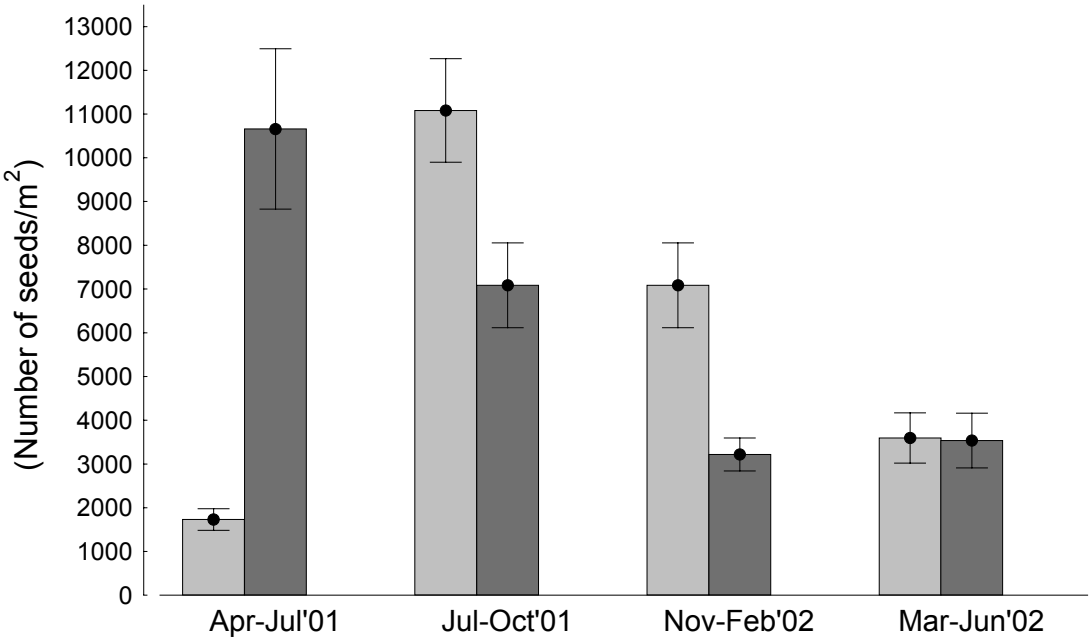

13000
12000
11000
10000
9000
8000
7000
6000
5000
4000
3000
2000
1000
0
(Number of seeds/m2)
Apr-Jul'01
Jul-Oct'01
Nov-Feb'02
Mar-Jun'02

Fig 4.-

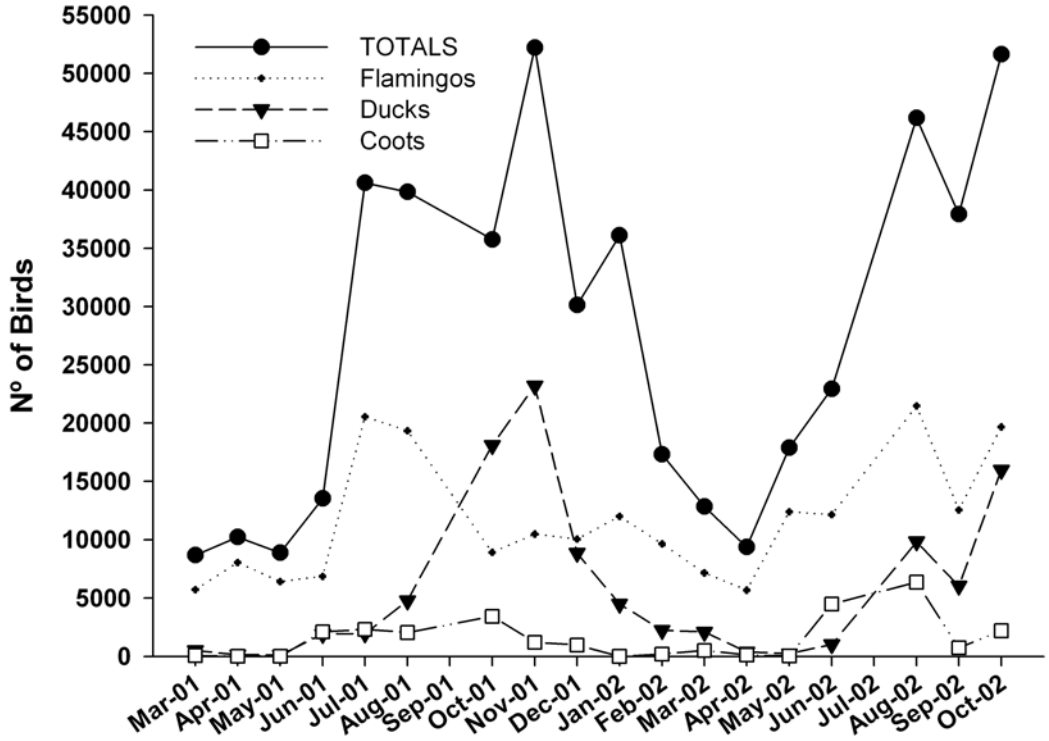

TOTALS
Flamingos
Ducks
Coots
Nº of Birds
55000
50000
45000
40000
35000
30000
25000
20000
15000
10000
5000
0
Mar-01
Apr-01
May-01
Jun-01
Jul-01
Aug-01
Sep-01
Oct-01
Nov-01
Dec-01
Jan-02
Feb-02
Mar-02
Apr-02
May-02
Jun-02
Jul-02
Aug-02
Sep-02
Oct-02

Fig 5.-

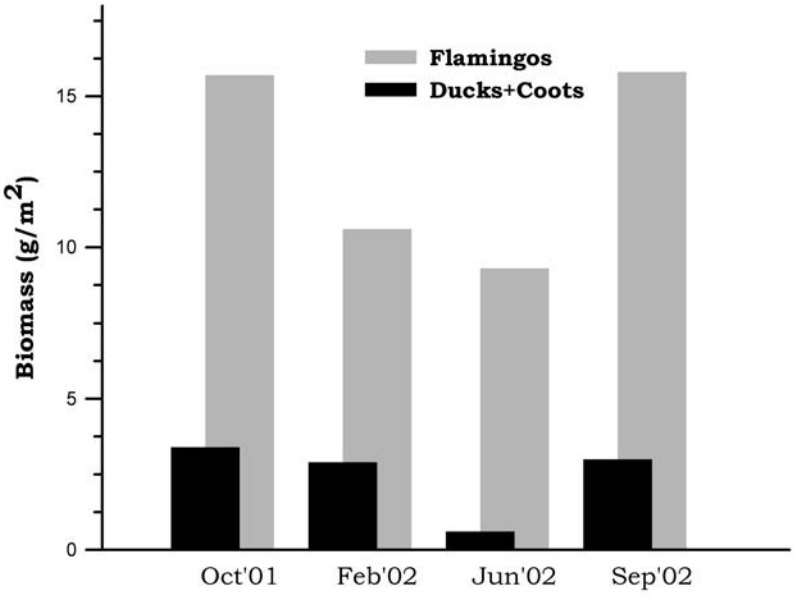

Flamingos
Ducks+Coots
Biomass (g/m2)
15
10
5
0
Oct'01
Feb'02
Jun'02
Sep'02

Fig 6.-

CAPÍTULO 2.

Efecto de las aves acuáticas sobre los invertebrados del bentos / Waterbirds effect on benthic invertebrates.

Basado en el manuscrito original: Rodríguez-Pérez, H., and Green, A.J. Strong effects of distinct waterbirds on a benthic community in shallow lakes, in different seasons throughout the year.

Resumen

El papel funcional de las aves acuáticas en los sistemas acuáticos ha sido habitualmente infravalorado en comparación con el de los peces. Hay poca información sobre los efectos de las aves acuáticas en los macroinvertebrados bentónicos en sistemas no mareales, con la excepción de los efectos sobre bivalvos. En una antigua marisma temporal, en el estuario del Guadalquivir, actualmente convertida en balsas de piscicultura extensiva, hemos comprobado los efectos de las aves acuáticas sobre los macroinvertebrados bentónicos, para ello hemos usado distintos tipos de cercados de exclusión de 3x3 metros, para excluir selectivamente a los flamencos comunes (*Phoenicopterus ruber*), o a todo el conjunto de aves acuáticas, y compara los resultados con cotroles. La duración de los experimentos fue de tres meses, y se repitieron cuatro veces a lo largo de un ciclo anual, considerando así las fluctuaciones estacionales en las densidades de aves e invertebrados. Hemos detectado fuertes efectos aditivos al excluir a los flamencos, y los patos y fochas en las variables de biomasa total del bentos y abundancia de larvas de quironómidos. En algunos casos ambos tratamientos, y en otros casos solamente uno de ellos, produjeron incrementos en la diversidad taxonómica, las abundancias de poliquetos y gasterópodos, y la masa media individual de poliquetos y gasterópodos. Se produjo una interacción positiva de Tratamiento x Perido para la variable masa seca total, y la diferencia más importante entre tratamientos en esta variable, ocurrió durante la invernada de los patos, durante la cual los controles sólo tuvieron un 25% de la mas seca total recogida en los cercados de exclusión de todas las aves acuáticas. No hemos encontrado efectos indirectos, y aparentemente la competencia tiene poca importancia como proceso regulador de este comunidad bentónica, comparada con el efecto producido por la depredación, la 'bioturbación' y la herbivoría de los patos. Nuestros resultados contradicen las sugerencias previas que aseguraban que los depredadores epibentónicos no tenían efectos importantes sobre el bentos en hábitats con vegetación sumergida. Las densidades de aves acuáticas registradas fueron relativamente bajas en comparación con las de otros trabajos, y nuestros resultados sugieren que los flamencos y los patos y fochas juegan un papel importante estructurando los lagos someros en la región mediterránea. El continuado incremento de las poblaciones de flamenco común en esta región probablemente tendrá consecuencias a nivel de ecosistema.

Palabras clave*:* cercados, comunidad del bentos, *bottom-up,* aves acuáticas, flamencos, *Phoenicopterus ruber*, especie claves, depredación, bioturbación.

Abstract

The functional role of waterbirds in aquatic systems has been largely neglected in comparison with the role of fish. With the exception of bivalves, there is little information on the effects of waterbirds on benthic macroinvertebrates in non-tidal habitats. In former temporary marshes converted into extensive fish ponds within the Guadalquivir estuary (SW Spain), we tested the effects of waterbirds on benthic macroinvertebrates using distinct exclosure designs to exclude either Greater Flamingos *Phoenicopterus ruber* or all waterbirds from 3x3m plots, and comparing them with controls. Experiments lasting three months were repeated four times during different periods of the annual cycle, with major differences in the relative densities of different invertebrate and bird groups. Strong positive additive effects of excluding flamingos and waterfowl (ducks and coots) were detected on the total biomass of benthos and the abundance of chironomid larvae. One or both exclosure treatments also increased taxon diversity, the abundance of polychaetes and gastropods, and the mean body mass of polychaetes and gastropods. There was a significant Treatment x Period interactions for total dry mass, and the difference between excluding flamingos and all birds was most marked during the waterfowl wintering period, when controls held only 25% of the dry mass recorded in waterfowl exclosures. We found no evidence of indirect effects, and competition may have little importance in structuring this benthic community compared with the effects of predation, bioturbation and herbivory by waterbirds. Our results contradict previous suggestions that epibenthic predators do not have strong effects on benthos in vegetated habitats. The densities of waterbirds present were relatively low compared with other studies, and our findings suggest that flamingos and waterfowl are of great importance in structuring shallow lakes in the Mediterranean region. Ongoing increases in numbers of Greater Flamingos in this region are likely to have consequences at the ecosystem level.

Keywords: exclosures, benthos community, bottom-up, waterbirds, flamingos, *Phoenicopterus ruber*, keystone, predation, bioturbation.

INTRODUCTION

Competition, predation and disturbance (both biotic and abiotic) are three major forces driving the structure of animal communities, but their relative strength depends on habitat type, environmental stress, vital rates, etc (Roughgarden and Diamond 1986, Menge and Sutherland 1987). In aquatic systems, many paradigms for community ecology have been established using intertidal rocky zones as a model system (Paine 1966, 1980, Lubchenco and Menge 1978, Sousa 1979, Menge et al 1994, Berlow 1997), but the relative importance of biological processes in structuring benthic soft-sediment communities appears to be different, owing to the fact that interspecific competition is weaker in this complex three dimensional habitat (Sih et al 1985,Wilson 1991, Olaffson et al 1994). While competition for resources is often the main determinant of community structure in other environments, predation seems to be the strongest determinant in soft-sediment communities (Peterson 1979, Wilson 1991, Olafsson et al 1994, Thrush 1999), where most studies of predation have focussed on invertebrates.

Predation is usually studied via exclosure experiments (see Sih et al 1985 for review). This experimental approach has its problems, but these can be overridden by correct replication (Hurlbert 1984) and setting of controls.

Waterbirds are important benthic predators both in marine and inland aquatic systems. Various exclosure studies have focussed on the role of shorebirds as predators on benthic invertebrates in the intertidal zone (Quammen 1984, Botto et al 1998), but studies of other bird groups or in other habitats are infrequent. It is well known that ducks often feed on benthic invertebrates (Kear 2005), and diving ducks are known to have profound effects on the abundance of bivalves (Hamilton et al 1994, Hamilton 2000, Mitchell et al 2000, Yamamuro 1998). However, there are few studies of the influence of waterfowl (Anatidae and coots) on other benthic invertebrates (Smith 1986, Bortolus et al. 1998, Marklund & Sandsten 2002). In contrast, the role of waterfowl as herbivores has been repeatedly investigated with exclosure experiments (Mitchell and Perrow 1998).

Exclosure experiments have shown that flamingos (Phoenocopteridae) have a significant effect on benthic organisms (Hurlbert and Chang 1983, Glassom and Branch a and b 1997) and on macrophytes (Rodríguez-Pérez and Green, 2006). These impacts of flamingos appear to be caused not only directly by predation, but also by the bioturbation produced by their feeding behaviour (Fig. 1). Greater Flamingos (*Phoenicopterus ruber*) have undergone a dramatic increase in population size in the Mediterranean region since 1970, and are now one of the most abundant waterbirds in this region in terms of biomass (Johnson 1997, Wetlands International 2002). We suggest that Greater Flamingos are

keystone species (*sensu* Power et al. 1996) in aquatic ecosystems, not only as predators but also as ecosystem engineers (see discussion).

Here we present an exclosure study comparing the effects of Greater Flamingos and waterfowl on benthic invertebrates in shallow brackish marshes in Doñana, south-west Spain, during distinct three month periods of the annual cycle. We consider the effects of the two waterbird groups on the abundance and size of different benthic organisms, as well as on community traits such as diversity, taxonomic richness and total dry mass. We show that predation by waterbirds is a major force structuring the benthic community and consider the effects of seasonal changes in bird and invertebrate abundance.

Ours is a novel study addressing simultaneously the impacts of two functionally different (*sensu* Duffy 2002) groups of waterbirds on a non-tidal benthos community (but see Marklund and Sandsten 2002), and comparing the effects at different times throughout the annual cycle (see Hamilton 2000 for such a study in an intertidal community).

Study area

We carried out exclosure experiments in Veta la Palma (36^{o}57'N, 6 ^{o}14'W), a private fish-farm divided in 52 regular ponds within Doñana Natural Park. The ponds were constructed in 1992-1993 on top of what was natural marshland in the Guadalquivir estuary (Fig. 2). This habitat has some similarities with marine soft sediments and is supplied with estuarine water. All the ponds are shallow (average 30 cm, maximum depth 50 cm) and flat-bottomed with a total combined surface area of 2997 ha. The fish species cultured are European Seabass (*Dicentrarchus labrax*), Flathead Mullet (*Mugil cephalus*), Gilthead seabream (*Sparus auratus*) and eels (*Anguilla anguilla*), as well as Atlantic ditch shrimp (*Palaemonetes varians*). The alien killifish *Fundulus heteroclitus* is also abundant, as is the exotic crab *Rhithropanopeus harrisii*. Each pond is dried out under rotation approximately every two years to extract fish. Ponds are interconnected via canals and permanent flow of water taken from the Guadalquivir estuary maintains dissolved oxygen levels (see Frisch et al. 2005 for more details).

The dominant submerged macrophyte is *Ruppia maritima*, forming extensive beds during spring and early summer. Small patches of *Potamogeton pectinatus* occur in some ponds in years of relatively low salinity. Most of the shoreline is bare mud and regular dredging to prevent siltation of peripheral canals used to extract fish from the ponds has restricted development of vegetation, which is dominated by *Arthrocnemum macrostachyum* and *Suaeda spp.* with some small patches of *Phragmites australis* and *Scirpus maritimus*. Mean monthly air temperature during the study varied from 11.8 ºC in February to 23.5 ºC in July. Mean annual precipitation is 562 mm/yr with a range of 158-1062 mm/yr (Castroviejo

1993). Salinity during our study varied from 7 g/l during winter months of high rainfall to 15 g/l at the end of September, after the dry summer months typical of the Mediterranean region. pH ranged from 9.3 to 10.4.

MATERIALS AND METHODS.

Aerial surveys of waterbirds in Veta la Palma were carried out monthly during the study as part of a wider survey program to census birds of the Doñana marshes. All birds were counted, including those on the water and in flight (most of which were flushed by the plane). Flamingos are consistently recorded in higher numbers in Veta la Palma than in surrounding areas whereas the opposite is true for ducks and coot (M.A. Rendon, A.J. Green and E. Aguilera, unpublished data). Eight ponds were used in each experimental period of 3 months. Three ponds were substituted when they were drained in order to extract fish, making a total of 11 different ponds used during the study (range 26-114 ha, making a total of 697 ha). We included ponds that had been flooded for different lengths of time when we started the study.

All exclosures and controls were 3 x 3m in size, and we used two exclosure treatments. One treatment used a 2 cm mesh nylon net tied to a square PVC frame suspended from four iron poles (2 m long, 10 mm diameter) pushed into the pond bottom in each corner. Wires were also attached from the top of the poles to the centre of the net to prevent it from touching the water surface. At the sides of the square frame, 20 cm of net hung vertically down to the water surface to prevent birds from passing underneath. This design allowed fish to enter but decreased utilisation by ducks, coot and other birds. The netting did not visibly affect water movement. There was no algal growth and shading effects were close to zero.

The second treatment excluded only flamingos by taking advantage of their extreme height. Four iron poles were placed in each corner of the square as before, and a wire tied round them at a height of 70-75 cm from the bottom (i.e. 30-65 cm above the water surface). This height was selected after field trials confirming that other birds passed comfortably underneath the wire and freely entered the exclosures. Controls consisted merely of the four iron poles. Eurasian coots *Fulica atra* and ducks were frequently observed feeding inside controls and flamingo exclosures, whereas birds were never observed inside all-birds exclosures. It is also possible that Black-tailed Godwit *Limosa limosa* and other larger shorebirds occasionally entered the shallowest plots.

Each experimental block contained one exclosure of each kind plus a control, separated by approximately 10 m (fig 2). Treatments were randomly located within each block. There were two experimental blocks in each pond, separated by approximately 25 m. Both blocks were placed in the western part of each pond, aligning all exclosures and

controls at an equal distance from the pond edge, so as to expose them equally to the dominant southwesterly wind with an equal and relatively low fetch. Benthos was sampled twice in each exclosure and control, both immediately after establishing the exclosures and three months after installing them. Four experiments of three months duration were performed between April 2001 and June 2002, moving the exclosures and controls 10 m in towards the centre of the pond at the beginning of each new experiment. There was no change in depth towards the centre of the ponds. Owing to the lack of a significant fringe of emergent vegetation, there was no windbreak along the pond edge and no gradient in waterbird density from the edge to the center of the ponds that could have influenced waterbird effects as we moved controls and exclosures between experimental periods (Weisner et al 1997).

The first experimental period ran from 3 April to 11 July 2001 (Jul 01 from hereon) coinciding roughly with the breeding period for waterbirds. The second period, from 13 July to 30 October 2001 (Oct 01 from hereon), corresponded with the post-breeding period. The third period from 22 November 2001 to 27 February 2002 (Feb 02 from hereon) covered the wintering period. Finally, the fourth period from 7 March to 6 June 2002 (Jun 02 from hereon) overlapped with the pre-breeding and early breeding periods. Each individual exclosure or control was set for 70-93 days in Jul 01, 82-92 days in Oct 01, 95-105 days in Feb 02 and 88-96 days in Jun 02. In a given pond and period, all exclosures and controls were established or sampled on the same day. There was a slight overlap between experimental periods caused by the time required to sample and move exclosures and controls in the ponds. This usually took two weeks, but bad weather caused some delays.

Three core samples, each of 23.8 cm^2 surface area, were taken on two occasions in each 3 x 3 m square plot. When plots were established, cores were taken from the inner side of the midpoint of three of the four sides so as to avoid stepping inside the plots. At the end of the three month period, the cores were taken from the central area at least 1 m from the edge, so as to reduce edge effects, while ensuring that each core was separated by at least 1 m from each other. The water depth at sampling points ranged from 10 to 39 cm (mean+s.e. = 27$\pm$8 cm), within the range accessible to dabbling ducks when feeding (Pöysä 1983). Invertebrates in the water column and submerged macrophytes were also sampled (Rodríguez-Pérez and Green 2006, authors unpublished data).

The upper 5 cm of sediments in each core was extracted with a PVC tube and fixed with formalin. Each sample was later sieved through a 250 μm mesh, then invertebrates were sorted into taxonomical groups and counted using a binocular microscope. We identified invertebrates to the highest practical taxonomic level (species level in some cases) using keys and help from specialists (see acknowledgments). To quantify the dry mass of each sample, invertebrates were dried for 24 hour at 80ºC and then weighed to the nearest

0.0001 g. This value was divided by the number of individuals counted to get an average mass for each taxon in each sample. When samples weighed less than the minimum value, half this value (i.e. a mass of 0.00005 g) was assumed.

Statistical methods

We analyzed the effects of different treatments (all-birds exclosures, flamingo exclosures, controls), experimental period and their interaction on the benthic invertebrate community using Generalized mixed Linear Models (GLMs, McCullagh and Nelder 1989). Dependent variables were counts (including zeros) and mean individual mass (as a body size estimate) of different taxa, as well as total sample dry mass, richness (number of taxa per sample) and a diversity index. We used a modified Simpson index (Dw) because it reflects the importance of both diversity and abundance (Sherfy and Kirkpatrick 2003). Data from the three cores taken in each plot were pooled prior to analysis. Pond and experimental block nested within ponds were included as random factors using the GLIMMIX procedure (SAS Institute 2005). Treatment and sampling period were included as fixed factors of three and four levels respectively, and the Treatment x Period interaction was included in the model when it was significant. *Post-hoc* analyses of least-squared means identified significant differences among levels of fixed factors, using Tukey-adjusted p-values. Tests on the effects of each predictor were performed using *F*-statistics (Crawley 1993). In all cases, we present models based on the data from the end of each experimental period. We analyzed data collected when installing the experimental plots separately, and in no case was there any significant difference in invertebrate abundance or community parameters among treatments (results not shown). Thus, we are confident that the differences observed among treatments three months later (see results) were not an artefact due to pre-existing patterns in invertebrate distribution.

All invertebrate groups recorded (see Table 8 for details) were considered for analysis, including invertebrates that are not strictly benthic. Count data were analyzed using a log link function and negative binomial error distribution (Gray 2005). Alternative dependent variables based on the difference recorded in the number of invertebrates from the beginning of an experimental period to the end were rejected because they included negative values to which the negative binomial error distribution can not be applied, and for which no suitable transformations could be found. Amphipods occurred in 37% of the samples, but models of counts for this and for rarer taxa did not converge. We thus only present models of counts for the 5 most abundant groups: polychaetes (excluding nereids), *Nereis diversicolor*, chironomid larvae, the gastropod *Potamopyrgus antipodarum* and the ostracod *Cyprideis torosa*.

We also present models of mean dry mass for these taxa (except *N. diversicolor*) as well as for the isopod *Lekansphaera hookeri* and the amphipod *Corophium orientale*. We preferred not to add *N. diversicolor* data to those for other polychaetes because they are so different ecologically. Nereids are much larger (Table 8), dig deeper burrows making them likely to be overlooked in our samples of 5 cm depth and, unlike other polychaetes, are predators (Verhoeven 1980, Smith et al 1996). Abundance and mass data for all taxa recorded were used in models of total dry mass, taxa richness and diversity. Taxa richness and diversity indices were calculated based on the finest taxonomical level reached, mixing species level with broader levels of identification.

Diversity (transformed as Dw^2), total dry mass per sample (log_{10} transformed) and mean dry mass per sample (log_{10} transformed) were analyzed using an identity link and normal error distribution, whereas taxa richness was analyzed using a log link and a poisson error distribution. Figures of invertebrate abundance, mass and richness are presented using geometric means calculated by log_{10} transformation of the raw data (adding 1 when necessary) calculating the arithmetic mean and SE and then back-transforming those outcomes with 10^x and plotting them, because they better represent the skewed data distributions in these cases.

RESULTS

Waterbird counts

Peak numbers of waterbirds in the study site occurred during the post-breeding period, reaching more than 50,000 in November 2001 (Fig. 3). Numbers were also high in winter, and were lowest in March and April (Fig. 3). Numbers of the largely herbivorous coots and omnivorous ducks followed a similar pattern, peaking in October-November during the post-breeding period. Numbers of flamingos reached a peak in July-August at the height of their breeding season (Fig. 3). In terms of biomass, flamingos were easily the most important group of birds in the area (Fig. 3). The mean density of ducks and coot in Veta la Palma was 2.6 ha^{-1}. The mean density of flamingos was 3.9 ind ha^{-1} (densities were calculated based on the total pond area of 2997 ha). The most abundant waterfowl species recorded (in order of decreasing abundance) were *Fulica atra, Anas platyrhynchos, A. clypeata, A. penelope, A. acuta, A. strepera, Netta rufina* and *Aythya ferina*. The *Anas* dabbling ducks accounted for 96% of the total number of ducks counted. *A. penelope*, *A. clypeata* and *A. acuta* were only present in winter.

Invertebrate fauna

Groups recorded in our core samples (Table 8) were the nereid polychaete *Nereis diversicolor*, other polychaetes (*Alkmaria romijni* and *Streblospio shrubsolli*), Turbellaria, the alien gastropod *Potamopyrgus antipodarum*, the anthurid isopod *Cyatura carinata*, the sphaeromatid isopod *Lekansphaera hookeri*, the amphipod *Corophium orientale*, the decapod *Palaemonetes varians*, the ostracod *Cyprideis torosa*, chironomid larvae, *Ephydra* sp. larvae, the corixid *Sigara stagnalis*, coleoptera larvae (*Berosus* sp. and *Enochrus* sp.) and nymphs of *Ischnura graellsi* (Odonata).

Measures of benthic community structure

Total sample mass was analysed as a measure of the standing crop of benthos. Treatment had a highly significant main effect on sample mass (Table 1, Fig. 4), with the highest mass in all-bird exclosures ("no birds"), lowest in controls and intermediate in flamingo exclosures ("no flamingos"). All pairwise effects between treatments were significant (Table 2, Fig. 4). There were also significant differences between periods, with Jun02 having significantly higher biomass than Feb02 (Table 2, Fig. 4). The Treatment x Period interaction was also significant, with Treatment effects being non-significant in Jul01 and Jun02(Table 7). Differences between all-birds exclosures and controls were highly significant in Oct01 and Feb02 (Table 7). The percentage reduction in arithmetic mean total dry mass attributed to "all birds" ranged from 23 to 75% across the four periods, and the reduction attributable to flamingos ranged from 7 to 48%. However, for both treatments, the first period Jul01 was the only one with reductions below 45 %.

Neither the Treatment nor Period main effects were significant in a model of taxonomic richness (Table 1), although richness was consistently lower in controls than in exclosures (Fig. 4).

In a model of diversity using a modified Simpson diversity index (weighted by abundance, Dw), Treatment and Period had highly significant effects (Table 1, Fig. 4). Dw was significantly higher in "no birds" than in controls (Table 2, Fig. 4), and was significantly higher in Jun02 than in other periods (Table 2, Fig. 4).

Invertebrate counts

N. diversicolor, other polychaetes*N. diversicolor,* the gastropod *P. antipodarum*, chironomid larvae and the ostracod *C. torosa* were the groups abundant enough to analyze with count data. Combined, these groups represented 92-98% of the total number of invertebrates and 83-97% of the total dry mass (range of arithmetic means for the four

experimental periods for samples taken at the beginning of each period). The treatment main effect was highly significant for non-nereid polychaetes, gastropods and chironomid larvae (Table 3). The Period main effect was highly significant for all taxa except other polychaetes (Table 3).

Numbers of other polychaetes were significantly higher in both exclosure treatments than in controls, and tended to be higher in “no birds” than “no flamingos”, although not significantly so (Table 4, Fig. 5). Numbers of chironomids were highest in “no birds” and lowest in controls, and all pairwise differences between treatments were significant (Table 4, Fig 5). Between periods, all pairwise differences were also significant, and the order of abundance was Oct01 > Jun02 > Feb02 > Jul01 (Table 4, Fig 5). Numbers of the snail *P. antipodarum* were significantly higher in “no birds” than in other treatments (Table 4, Fig 5). They were significantly higher in Jul01 than in other periods (Table 4, Fig 5).

Numbers of the ostracod *C. torosa* were significantly higher in Jun02 than in any other period, and significantly higher in Feb02 than Oct01 (Table 4, Fig 5). Numbers of *N. diversicolor* were significantly higher in Jul01 than in Feb02 and Jun02 (Table 4, Figure 5). Although the treatment effect was not significant, numbers were consistently lower in controls than in exclosures (Fig. 5).

Invertebrate size (mass)

We analyzed the mean mass per sample for four of the five taxa *N. diversicolor*whose numbers we analyzed (*N. diversicolor* did not converge), as well as the isopod *Lekanesphera hookeri* and the amphipod *Corophium orientale*. Treatment had a significant effect on the mass of polychaetes and gastropods, but not chironomids, ostracods, isopods or amphipods (Table 5). The Period main effect was highly significant for all six taxa, whereas there were no significant Treatment x Period interactions (Table 5).

The mean mass of polychaetes was significantly higher for “no birds” than for controls, and was significantly lower for Feb02 than for other periods (Table 6, Fig 6). The mean mass of *P. antipodarum* was significantly higher in both exclosure treatments than in controls, and there was a non-significant trend to be larger in “no birds” than “no flamingos” (Table 6, Fig 6). The mean mass was significantly higher in Jun02 than in Oct01 and Feb02 (Table 7, Fig 6).

For chironomid larvae, all pairwise differences between periods were significant with the exception of Oct01 vs Jun02, the order of mean mass being Feb02 > Jul01 > Jun02 > Oct01. The mean mass of the ostracod *C. torosa* was significantly higher in Jun02 (when they were most abundant) than Jul01 and Feb02 (Table 6, Fig 6). The mean mass of the isopod *L. hookeri* was significantly lower in Feb02 (when they were least abundant) than in

Jul01 and Jun02 (Table 7, Fig 6). The mean mass of the amphipod *C. orientale* was significantly lower in Oct01 than in Feb02 (Table 6, Fig 6).

DISCUSSION

Our experiments suggest that flamingos and other waterbirds (essentially ducks and coots) have profound and additive effects on the benthic invertebrate community. Total dry mass and chironomid abundance were increased by excluding flamingos, and more so by excluding all waterbirds. Diversity and the size and abundance of polychaetes and gastropods were also increased by excluding waterbirds. Our results indicate that predation by waterbirds is a major force influencing the structure of the benthic community in our study area.

Taxa richness was consistently increased by exclusion (Fig. 4), and we suggest that the treatment effect was non-significant owing to a scale effect. The larger the sample size, the more taxa are likely to be recorded. In our models, we summed three samples taken from each plot because they were not spatially independent. Differences in taxa richness between treatments were more pronounced at the sample scale (not shown) than at the plot scale (Fig. 4). The fact that three taxa (chironomids, snails and other polychaetes) were significantly more abundant in exclosures also indicates that, at a fine scale, richness is also increased. Furthermore, given our incomplete identification of taxa, our results are likely to underestimate the effects of waterbirds on species richness.

Greater flamingos are known to consume polychaetes, ostracods, chironomid larvae and other benthos (Glassom & Branch 1997 a, Johnson 1997, Rodríguez-Pérez and Green 2006). In our study area, analysis of the gut contents of ducks and coot has confirmed that they consume chironomid larvae, isopods, polychaetes, *C. torosa* and *P. antipodarum* (Green & Sánchez 2003, authors unpublished data). However, our results are also likely to reflect the bioturbation (*sensu* Reise 2002) effects of waterbirds and the effects of exclosure on submerged macrophytes (see below), which themselves have a major structuring role on invertebrate communities (see Jeppesen et al 1998 for review).

Experiments such as ours, in which predators are excluded, often have indirect or unexpected effects in which exclusion decreases the density of some species, or changes the size distribution of a given species in a manner that cannot be explained directly via predation by the excluded predators (see Sih et al. 1985, Wooton 2002 for review). Such results have been explained either by compensatory predation in which a secondary predator increases its abundance in exclosures (Kneib 1988, Thrush et al 1994, Hamilton 2000), or by interspecific or adult-larval intraspecific competition (Peterson 1979,Wilson 1991, Thrush 1999). Adult-larval competition is thought to play an important role in soft sediment

communities, and typically leads to a reduction in density but increase in size in exclosures (Woodin 1976, Peterson 1979, Wilson 1991, Olafsson et al 1994). In contrast, we did not find evidence for any taxa of a decrease in abundance in exclosures. In general, we found exclusion to increase both the abundance and mean size (mass) of invertebrates.

Our results thus suggest that competition may have relatively little influence on community structure in our study area, and support Sih et al.'s (1985) prediction that indirect effects are more common when predation intensity is low. However, we cannot rule out the possibility that indirect effects would have been manifested if we had maintained exclosures for a longer time (see Hamilton 2000). In the community studied, the benthos is dominated by detritivores feeding directly on organic matter or on the epibenthic algae and diatoms that grow on it. However, there is little information available on the extent of interspecific competition between polychaetes, chironomid larvae, ostracods and the other groups recorded. Polychaetes tend to use inner layers of sediments whereas chironomids use outer layers, thus reducing the scope for competition between them (Angradi et al 2001).

Animal-mediated sediment disturbance can have significant deleterious effects on suspension-feeders and tube-builders such as polychaetes and chironomid larvae in infaunal communities (Wilson 1991). Crabs, fish, shorebirds and flamingos are all known to cause significant levels of disturbance to soft-sediment communities (Quammen 1984, Thrush et al 1994, Glassom and Branch 1997a,1997b). In our study area, flamingos form craters while feeding (Fig. 1) and uproot the submerged macrophyte *Ruppia maritima* (Rodríguez-Pérez and Green, 2006). Waterfowl are also likely to cause disturbance to sediments while grazing on *R. maritima* and feeding on benthic invertebrates. Submerged vegetation itself creates a refuge from sediment disturbance and from predation (Woodin 1978, Peterson 1979, Beckett 1992 , Olafsson et al 1994). Since we have even found effects of waterbirds on benthos in summer when *R. maritima* is at peak biomass (Rodríguez-Pérez and Green, 2006), our results contradict Peterson's (1979) conclusion (still widely accepted, Olaffson et al 1994, Thrush 1999) that epibenthic predators do not have strong effects on benthos in vegetated habitats.

Intermediate levels of disturbance are generally expected to increase community diversity by limiting the abundance of dominant species (Sousa 1979, Paine and Levin 1981). By excluding a disturbance factor (waterbirds), we found that diversity increased suggesting that, in our study system, either disturbance levels are very high (e.g. owing to the effects of wave action, fish, etc, see Olaffson et al 1994) or that predation effects are much more important than disturbance effects. However, the results of analyses using diversity indices are heavily dependent on the precise index used. Although our "no birds" exclosures may conceivably have had a slight influence on hydrodynamics and sedimentation due to slight contact between the net and the water surface, we have no

evidence of this. There was no difference between controls and flamingo exclosures in their possible effects on water flow or sedimentation, yet the exclusion of flamingos had a profound effect on the benthos, reducing the abundance of polychaetes and chironomids and the size of gastropods.

The great majority of previous work investigating the influence of waterbirds on benthic invertebrates has focussed on the influence of seasonal concentrations of shorebirds in tidal mudflats (e.g. Quammen 1984, Botto et al 1998) and other soft sediments (Székely and Bemberger 1992, Sánchez and Green in press) during migration or wintering periods. Many of these studies have found significant effects of shorebirds, but others have suggested that the exclusion of birds has no effect owing to compensation by other predators such as other invertebrates (Marsh 1986). With the exception of studies of the effects of diving ducks on bivalves (Hamilton et al 1994, Hamilton 2000, Mitchell et al 2000), ours is one of few exclosure experiments to study the impact of waterfowl on benthos. Smith et al. (1986) and Ashley et al (2000) found no influence of waterfowl on the abundance of dipteran larvae, and Marklund et al (2002) found no negative effects of waterfowl on total macroinvertebrate abundance and biomass (including non-benthos). Others have shown herbivorous waterfowl to reduce the abundance of epiphytic and benthic invertebrates at those times of the year when the density of waterfowl peaks (Bortolus et al. 1998, Marklund & Sandsten 2002, Sherfy and Kirkpatrick 2003).

Our study provides the strongest evidence to date that waterbirds have a major influence on benthic invertebrate abundance and community structure in shallow lakes (*sensu* Scheffer 1998) throughout the annual cycle, and not just at times of peak migratory concentrations. Exclosure effects were apparent throughout the annual cycle, and the only model with a significant Treatment x Period interaction was the one for total dry mass. This interaction suggested that the difference between controls and “no birds” was stronger in Oct01 and Feb02, periods when waterfowl (ducks and coot) were relatively more abundant and when the combined abundance of waterfowl and flamingos was highest (Table 7, Fig 3). However, exclosure effects are also likely to depend critically on how the timing of the experiment relates to growth rates and other aspects of invertebrate life cycles (Mitchell and Wass 1996), as well as on diet switches by waterbirds in relation to in their own nutritional needs and changes in the size distribution of available prey (Krapu & Reinecke 1992, Kear 2005). For example, invertebrate growth, dispersal and reproductive rates might be expected to be lower in Feb02 when water temperatures were the lowest, and the same level of predation during this period might produce a stronger exclosure effect.

In a metanalysis strongly dependent on studies of diving ducks feeding on bivalves, Marklund et al. (2002) found a strong correlation between reduction of invertebrates and waterfowl density. However, at densities of waterfowl as low as ours (see results), they found

no evidence of impacts on benthic macroinvertebrates. The strength of the effects we recorded is even more significant bearing in mind the high density of fish in the study area. The fish present are likely to compete with waterbirds for benthic food (Cardona et al. 2001, Richardson et al 1990), and compensatory predation by fish may even have led us to underestimate the influence of birds on benthos (Marklund et al 2002).

The Greater Flamingo has previously been shown by exclosure experiments to influence the macro- and meiofauna in intertidal areas (Glassom and Branch 1997a and b), whereas the Andean flamingo (*Phoenicoparrus andinus*) also has a profound effect on microbenthos (Hurlbert and Chang 1983). The Greater Flamingo can be considered a bioturbator because it alters the sediment topography (Fig. 1, Kvitek et al 1992, Thrush et al 1994) and modifies physico-chemical sediment features such as redox potential and organic content (Glassom and Branch 1997b). Such sediment features are closely related to the densities of benthic fauna (Marinelli and Woodin 2002). Such bioturbation by macrofauna can even affect biogeochemical cycles (Lohrer et al 2004), and the Greater Flamingo is arguably a keystone species (*sensu* Power et al. 1996) in Mediterranean marshes (see also Rodríguez-Pérez and Green 2006). It has even been suggested that the modern absence of flamingos from salt lakes in Australia has had a strong influence on the fauna there (Bayly 1993). Numbers of Greater Flamingos are continuing to increase in the Mediterranean region due to management measures focussed on this particularly attractive bird (Wetlands International 2002). Given our results, this increase is likely to have consequences at the ecosystem level which should be taken into account in management decisions.

The total biomass of invertebrates recorded in our controls was much higher in the summer periods, probably reflecting the general increase in invertebrate production rates between April and June. However, it is striking how the biomass in all-bird exclosures was similar between periods (Fig. 4). This suggests the sediments may have a maximum invertebrate abundance that can be reached at any time of the year in the absence of birds, and further indicates that birds have a profound effect on this system. Peterson (1979) suggested that this maximum abundance would be limited by adult-larval interactions. However, although we found many significant differences between times of the year in the abundance and mass of invertebrates, these may not be seasonal effects *per se*. Since we studied active fish ponds that had been reflooded before starting the experiments, seasonal changes may have been confounded with a succession in the invertebrate communities as the ponds matured.

Non-nereid polychaetes were the most abundant benthic group both numerically and in terms of biomass (Table 8). Exclusion of flamingos and all birds had similar effects on their numbers, suggesting that flamingos were the bird group having the most impact on polychaetes. In tidal lagoons in Namibia, exclusion of flamingos increased the abundance of

three polychaete species, but the largest species *Diopatra neapolitana* was more abundant in controls (Glassom & Branch 1997a). However, differences in mean mass suggested that waterfowl reduced this size of non-nereid polychaetes more than flamingos. Shorebirds were previously reported to change the size distribution of polychaetes in tidal mud flats (Botto et al 1998).

The great importance of chironomid larvae in waterfowl diets is well known (Krapu & Reinecke 1992, Kear 2005). This was the only taxon for which all three treatments had significantly different numbers of larvae, and the difference between “no flamingos” and “no birds” suggests that waterfowl had relatively more effects on chironomids than on polychaetes. This may be partly because chironomids are epifaunal and polychaetes infaunal (Kornijow and Moss 1998, Angradi et al 2001), whilst flamingos feed deeper in the sediments via their trampling method that produces craters. Benthic chironomids may also be more negatively affected than polychaetes by the loss of macrophytes caused by waterfowl herbivory (Marklund & Sandsten 2002, Sherfy and Kirkpatrick 2003, but see Wrubleski 1989).

Numbers of *P. antipodarum* were only increased significantly by excluding all birds, suggesting that waterfowl were the major predators on these invertebrates. Gastropods can be very important in the diet of ducks (Krapu & Reinecke 1992). However, excluding flamingos increased the mean mass of *P. antipodarum*, suggesting that they too had important impacts on this snail. Since waterbirds reduce the amount of *R. maritima* and these snails are likely to feed on this plant or its epiphytes (Talbot and Ward 1987), the changes in snail abundance or size may potentially be mediated via changes in food supply, which influence growth or recruitment rates. Unlike another study of this species in its introduced range (Schreiber et al. 1998), we found the size of *P. antipodarum* to be highest in summer.

In conclusion, we have found unique evidence for strong top-down control of benthos by waterbirds in shallow lakes. This strong functional role of waterbirds in aquatic ecosystems has been largely overlooked, in contrast with the extensive literature on such a role for fish (Green and Figuerola 2005). The nature of this structuring role is complex, with major differences between waterbird groups and seasons which are not entirely consistent between different benthic taxa. Future studies are required to determine to what extent this structuring role of birds is consistent between systems. For example, how does it vary with hydroperiod and is it maintained in the temporary systems that abound in semi-arid and arid environments? To what extent is the observed role for birds dependent on predation and to what extent on bioturbation and herbivory? Future research should also establish whether indirect effects mediated via competition or compensatory predation would become apparent in longer term exclosure experiments.

Acknowledgements.

Maria del Mar Delgado provided invaluable assistance processing samples. We also thank Esther Macarro for field assistance, and Jordi Figuerola, Carlos M. Herrera, Pedro Jordano and Antonio Manzaneda for statistical advice. Ángel Baltanás, Arnthor Gardarsson, José Manuel Guerra, Andrés Millán and Juan Carlos Pérez assisted with taxonomic identifications. Pesquerías Isla Mayor S.A. and the Department of Environment (Consejería de Medio Ambiente) of the Andalusian Autonomous Government gave us the permission to work in Veta la Palma. This work was funded by the Consejería de Medio Ambiente. Héctor Rodríguez Pérez had a PhD grant from the CSIC-I3P programme funded by the European Social Fund. Aerial count data and the aerial photograph were provided by the Héctor Garrido and the Equipo de Seguimiento de Procesos of Doñana Biological Station.

References:

Angradi, T. R., S. M. Hagan, and K. W. Able. 2001. Vegetation type and the intertidal macroinvertebrate fauna of a brackish marsh: Phragmites vs. Spartina. Wetlands 21:75-92.

Argano, R. 1979. Guide per il riconoscimiento delle specie animali delle acque interne italiane 5. Isopodi. Consiglio Nazionale delle Ricerche.

Ashley, M. C., J. A. Robinson, L. W. Oring, and G. A. Vinyard. 2000. Dipteran standing stock biomass and effects of aquatic bird predation at a constructed wetland. Wetlands 20:84-90.

Askew, R.R. 1988. The dragonflies of Europe. 291 pp. Harley Books.

Baldó, F., M. A. Arias, and P. Drake. 2001. La comunidad macrobentónica del estuario del Guadalquivir. Boletín del Instituto Español de Oceanografía 17:137-148.

Bayly, I.A.E. 1993. The fauna of athlassic saline waters in Australasia and the Altiplano of South-America- Comparisons and historical perspectives. Hydrobiologia 267(1-3):225-231.

Beckett, D. C., T. P. Aartila, and A. C. Miller. 1992. Contrasts in density of benthic invertebrates between macrophyte beds and open littoral patches in Eau Galle Lake, Wisconsin. American Midland Naturalist 127:77-90.

Berlow, E.L. 1997. From canalisation to contingency: historical effects in a successional rocky intertidal community. Ecological Monographs 67:435-460.

Bortolus, A., O. O. Iribarne, and M. M. Martinez. 1998. Relationship between waterfowl and the seagrass Ruppia maritima in a southwestern Atlantic coastal lagoon. Estuaries 21:710-717.

Botto, F., O. O. Iribarne, M. M. Martinez, K. Delhey, and M. Carrete. 1998. The effect of migratory shorebirds on the benthic species of three southwestern Atlantic Argentinean estuaries. Estuaries 21:700-709.

Cardona, L., P. Royo, and X. Torras. 2001. Effects of leaping grey mullet Liza saliens (Osteichthyes, Mugilidae) in the macrophyte beds of oligohaline Mediterranean coastal lagoons. Hydrobiologia 462:233-240.

Castello, J. 1986. Contribución al conocimiento biológico de los crustáceos del litoral Catalano-Balear. PhD Thesis. Barcelona University.

Castroviejo, J. 1993. Mapa del Parque Nacional de Doñana. Consejo Superior de Investigaciones Científicas y Agencia de Medio Ambiente de la Junta de Andalucía, Madrid.

Crawley, MJ. 1993. GLIM for ecologist. Blackwell Science.

Del Hoyo, J., Elliot, A. and Sargatal, J. (eds.). 1992. Handbook of the birds of the world. - Lynx Editions.

Duffy, J. E. 2002. Biodiversity and ecosystem function: the consumer connection. Oikos 99:201-219.

Fauvel, P. 1923. Faune de France 5. Polychètes Errantes. Le Chavalier, Paris.

Fauvel, P. 1927. Faune de France 16. Polychètes Sédentaires. Le Chevalier, Paris.

Frisch, D., H. Rodríguez-Pérez, and A.J. Green. In press. Invasion of artificial ponds in Doñana Natural Park, southwest Spain, by an exotic estuarine copepod. Aquatic conservation: Marine and Freshwater ecosystem. DOI: 10.1002/aqc.718.

Ghetti, P.F., and K. Mc Kenzie. 1981. Guide per il riconoscimiento delle specie animali delle acque interne italiane 11. Ostracodi. Consiglio Nazionale delle Ricerche.

Glassom, D., and G. M. Branch. 1997a. Impact of predation by greater flamingos Phoenicopterus ruber on the macrofauna of two southern African lagoons. Marine Ecology Progress Series 149:1-12.

Glassom, D., and G. M. Branch. 1997b. Impact of predation by greater flamingos Phoenicopterus ruber on the meiofauna, microflora, and sediment properties of two southern African lagoons. Marine Ecology Progress Series 150:1-10.

Gray, B.R. 2005. Selecting a distributional assumption for modelling relative densities of benthic macroinvertebrates. Ecological Modelling 185(1):1-12.

Green, A.J., and J. Figuerola. 2005. Recent advances in the study of long-distance dispersal of aquatic invertebrates via birds. Diversity and Distributions 11(2):289-296.

Green, A.J., and M.I. Sánchez. 2003. Spatial and temporal variation in the diet of Marbled Teal Marmaronetta angustirostris in the western Mediterranean. Bird Study 50:153-160.

Hamilton, D. J. 2000. Direct and indirect effects of predation by Common Eiders and abiotic disturbance in an intertidal community. Ecological Monographs 70:21-43.

Hamilton, D. J., C. D. Ankney, and R. C. Bailey. 1994. Predation of Zebra Mussels by Diving Ducks - an Exclosure Study. Ecology 75:521-531.

Holthe, T. 1986. Polychaeta Terebellomorpha. Marine Invertebrate of Scandinavia 7. Norwegian University Press, Oslo.

Hurlbert, S. H. 1984. Pseudoreplication and the design of ecological field experiments. Ecological Monographs 54:187-211.

Hurlbert, S. H., and C. C. Y. Chang. 1983. Ornitholimnology - Effects of Grazing by the Andean Flamingo (Phoenicoparrus-Andinus). Proceedings of the National Academy of Sciences of the United States of America-Biological Sciences 80:4766-4769.

Jansson, A. 1986. The corixidae (Heteroptera) of Europe and some adjacent regions. 94 pp. Acta Entomologica Fennica 47.

Jeppensen, E., M. Sondergaard, M. Sondergaard, and K. E. Christoffersen. 1998. The structuring role of submerged macrophytes in lakes. Springer-Verlag, New York.

Johnson, A. R. 1997. Long-term studies and conservation of Greater Flamingos in the Camargue and Mediterranean. Colonial Waterbirds 20:306-315.

Kear, J. (Ed.). 2005. Ducks, geese and swans. Oxford University Press, Oxford.

Kneib, R. T. 1988. Testing for Indirect Effects of Predation in an Intertidal Soft-Bottom Community. Ecology 69:1795-1805.

Kornijow, R, and B. Moss. 1998. Vertical distribution of in-benthos in relation to fish and floating-leaved macrophyte population. Pages 227-232 in E. Jeppensen, M. Sondergaard, M. Sondergaard, and K. E. Christoffersen. 1998. The structuring role of submerged macrophytes in lakes. Spring-Verlag, New York.

Krapu, G.L., and K.J. Reinecke. 1992. Foraging ecology and nutrition. Pages 1-29 in Batt, D.B.J., A.D. Afton, M.G. Anderson, C.D. Ankney, D.H. Johnson, J.A. Kadlec, and G.L. Krapu (eds.) Ecology and mangement of breeding waterfowl. University of Minnesota Press, Mineapolis and London.

Kvitek, R. G., J. S. Oliver, A. R. Degange, and B. S. Anderson. 1992. Changes in Alaskan Soft-Bottom Prey Communities Along a Gradient in Sea Otter Predation. Ecology 73:413-428.

Lohrer, A. M., S. F. Thrush, and M. M. Gibbs. 2004. Bioturbators enhance ecosystem function through complex biogeochemical interactions. Nature 431:1092-1095.

Lubchenco, J., and B.A. Menge. 1978. Community development and persistence in low rocky intertidal zone. Ecological Monographs 59:67-94.

Marinelli, R. L., and S. A. Woodin. 2002. Experimental evidence for linkages between infaunal recruitment, disturbance, and sediment surface chemistry. Limnology and Oceanography 47:221-229.

Marklund, O., and H. Sandsten. 2002. Reduction of benthic macroinvertebrates due to waterfowl foraging on submerged vegetation during autumn migration. Aquatic Ecology 36:541-547.

Marklund, O., H. Sandsten, L.-A. Hansson, and I. Blindow. 2002. Effects of waterfowl and fish on submerged vegetation and macroinvertebrates. Freshwater Biology 47:2049-2059.

Marsh, C. P. 1986. Rocky Intertidal Community Organization - the Impact of Avian Predators on Mussel Recruitment. Ecology 67:771-786.

Mc Cullagh, P. and Nelder, J.A. 1989. Generalized linear models. Chapman and Hall.

Menge, B. A., and J. P. Sutherland. 1987. Community Regulation - Variation in Disturbance, Competition, and Predation in Relation to Environmental-Stress and Recruitment. American Naturalist 130:730-757.

Menge, B.A., E.L. Berlow, C.A. Blanchette, S.A. Navarrete, and S.B. Yamada. 1994. The keystone species concept: variation in interaction strength in a rocky intertidal habitat. Ecological Monographs 64:249-286.

Mitchell, J.S., R.C. Bailey, and R.W. Knapton. 2000. Effects of predation by fish and wintering ducks on dreissenid mussels at Nanticoke, lake Eire. Ecoscience 7:398-409.

Mitchell, S.F. and Perrow, M.R. 1998. Interaction between grazing birds and macrophytes. Pages 175-196 in E. Jeppensen, Sondergaard, M., Sondergaard, M. and Christoffersen, K.E., editors. The structuring role of submerged macrophytes in lakes. - Spring-Verlag.

Mitchell, S.F. and R.T. Wass. 1996. Quantifying herbivory: grazing consumption and interaction strenght. Oikos 76:573-576.

Olafsson, E. B., C. H. Peterson, and W. G. Ambrose. 1994. Does Recruitment Limitation Structure Populations and Communities of Macroinvertebrates in Marine Soft Sediments - the Relative Significance of Presettlement and Postsettlement Processes. Oceanography and Marine Biology, 32:65-109.

Ortiz, M., and A. Jimeno. 2001. Guía ilustrada para la identificación de las familias y los géneros de los anfípodos del suborden gammaridea de la península Ibérica. Graellsia 57(2):3-93.

Paine, R.T. 1966. Food web complexity and species diversity. American Naturalist 100:65-75.

Paine, R.T. 1980. Food webs: linkage, interacrion strength, and community infrastructure. Journal of Animal Ecology 49:667-685.

Paine, R.T., and S.A., Levin. 1981. Intertidal landscapes: disturbance and the dynamics of pattern. Ecological Monographs 51(2):145-178.

Peterson, C. H. 1979. Predation, Competitive exclusion, and diversity in the soft-sediment benthic communities of estuaries and lagoons.233-264.

Power, M. E., D. Tilman, J. A. Estes, B. A. Menge, W. J. Bond, L. S. Mills, G. Daily, J. C. Castilla, J. Lubchenco, and R. T. Paine. 1996. Challenges in the quest for keystones. Bioscience 46:609-620.

Pöysä, H. 1983. Morphology-mediated niche organization in a guild of dabbling ducks. Ornis Scandinavica 14:317-326.

Quammen, M. L. 1984. Predation by Shorebirds, Fish, and Crabs on Invertebrates in Intertidal Mudflats - an Experimental Test. Ecology 65:529-537.

Reise, K. 2002. Sediment mediated species interactions in coastal waters. Journal of Sea Research, 48, 127-141.

Richardson, W. B., S. A. Wickham, and S. T. Threlkeld. 1990. Foodweb Response to the Experimental Manipulation of a Benthivore (Cyprinus-Carpio), Zooplanktivore (Menidia-Beryllina) and Benthic Insects. Archiv Fur Hydrobiologie 119:143-165.

Rodríguez-Pérez, H., and A.J. Green. 2006. Waterbird impacts on widgeongrass Ruppia maritime in a Mediterranean wetland: comparing bird groups and seasonal effects. Oikos 112: 525-534.

Roughgarden, J., and J. Diamond. 1986. Overview: The role of species interactions in community ecology. Pages 333-343 in Community Ecology. Jared Diamond and Ted J. Case. 665 pp. Harper & Row Publishers Inc.

Ruffo, S. 1982. The Amphipoda of the Mediterranean. Mémoires de l'Institut Océanographique 13. Fondation Albert I Prince de Monaco.

Sánchez, M.I., and A.J. Green. In press. Shorebird predation affects density, biomass and size distribution of benthic chironomids in salt pans: an exclosure experiment. Journal of North American Benthological Society.

SAS Institute Inc. 2005. SAS GLIMMIX.

Scheffer, M. 1998. Ecology of shallow lakes. 357 pp. Kluwer Academic Publisher, Dordrecht.

Schreiber, E. S. G., A. Glaister, G. P. Quinn, and P. S. Lake. 1998. Life history and population dynamics of the exotic snail Potamopyrgus antipodarum (Prosobranchia : Hydrobiidae) in Lake Purrumbete, Victoria, Australia. Marine and Freshwater Research 49:73-78.

Sherfy, M. H., and R. L. Kirkpatrick. 2003. Invertebrate response to snow goose herbivory on moist-soil vegetation. Wetlands 23:236-249.

Sih, A., P. Crowley, M. Mcpeek, J. Petranka, and K. Strohmeier. 1985. Predation, Competition, and Prey Communities - a Review of Field Experiments. Annual Review of Ecology and Systematics 16:269-311.

Smith, D., R. G. Hughes, and E. J. Cox. 1996. Predation of epipelic diatoms by the amphipod Corophium volutator and the polychaete Nereis diversicolor. Marine Ecology Progress Series 145:53-61.

Smith, L. M., L. D. Vangilder, R. T. Hoppe, S. J. Morreale, and I. L. Brisbin. 1986. Effect of diving ducks on benthic food resources during winter in South Carolina, U.S.A. Wildfowl 37:136-141.

Sousa, W.P. 1979. Disturbance in marine intertidal boulder fields. The nonequilibrium maintenace of species diversity. Ecology 60(6):1225-1239.

Székely, T., and Z. Bamberger. 1992. Predation of waders (Charadrii) on prey populations: an exclosure experiment. Journal of Animal Ecology 61:447-456.

Tachet, H., P. Richoux, M. Bournaud and P. Usseglio-Polaterra. 2003. Invertébrés d'eau douce- Systématique, biologie, ecologie. 587 pp. CNRS Editions, Paris.

Talbot, J.M., and J.C. Ward. 1987. Macroinvertebrates associated with aquatic macrophytes in lake Alexandrina, New Zealand. Journal of Marine and Freswater Research 21:199-213.

Thrush, S. E. 1999. Complex role of predators in structuring soft-sediment macrobenthic communities: Implications of changes in spatial scale for experimental studies. Australian Journal of Ecology 24:344-354.

Thrush, S. F., R. D. Pridmore, J. E. Hewitt, and V. J. Cummings. 1994. The Importance of Predators on a Sandflat - Interplay between Seasonal-Changes in Prey Densities and Predator Effects. Marine Ecology-Progress Series 107:211-222.

Verhoeven, J. T. A. 1980. The Ecology of *Ruppia*-Dominated Communities in Western-Europe .2. Synecological Classification - Structure and Dynamics of the Macroflora and Macrofauna Communities. Aquatic Botany 8:1-85.

Weisner, S. E. B., J. A. Strand, and H. Sandsten. 1997. Mechanisms regulating abundance of submerged vegetation in shallow eutrophic lakes. Oecologia 109:592-599.

Wetlands International. 2002. Waterbird population estimates. - Wetlands International.

Wilson, W. H. 1991. Competition and Predation in Marine Soft-Sediment Communities. Annual Review of Ecology and Systematics 21:221-241.

Woodin, S. A. 1976. Adult-Larval Interactions in Dense Infaunal Assemblages - Patterns of Abundance. Journal of Marine Research 34:25-41.

Woodin, S. A. 1978. Refuges, Disturbance, and Community Structure - Marine Soft-Bottom Example. Ecology 59:274-284.

Wootton, J. T. 2002. Indirect effects in complex ecosystems: recent progress and future challenges. Journal of Sea Research 48:157-172.

Wrubleski, D. A. 1989. The effect of waterfowl feeding on a chironomid (Diptera: Chironomidae) community. Pages 691-696 in R. R. Sharitz and J. W. Gibbons, editors. Freshwater wetlands and wildlife. US Department of Environment Office of Scientific and Technical Information, Oak Ridge, Tennessee.

Yamamuro, M., Oka, N. & Hiratsuka, J. 1998. Predation by diving ducks on the biofouling mussel *Musculista senhousia* in a eutrophic estuarine lagoon. Marine Ecology-Progress Series, 174, 101-106.

FACTOR	Total weight						Richness						Diversity (Dw)					
	Estimate	SE	df_N	df_D	F	p	Estimate	SE	df_N	df_D	F	p	Estimate	SE	df_N	df_D	F	p
Intercept	-1.16	0.08					1.73	0.82					2.74	0.19				
Treatment (T)			2, 159		23.78	<0.0001			2, 165		1.69	0.2			2, 165		4.43	<0.0001
C	-0.26	0.1					-0.14	0.08					-0.44	0.15				
F	-0.02	0.1					-0.08	0.08					-0.29	0.15				
Date (D)			3, 159		3.45	0.02			3, 165		1.07	0.4			3, 165		7.66	<0.0001
Jul 01	-0.07	0.1					0.05	0.09					-0.62	0.19				
Oct 01	-0.03	0.1					-0.06	0.09					-0.8	0.18				
Feb 02	0.04	0.1					-0.1	0.09					-0.63	0.18				
TxD			6, 159		2.29	0.04						n.s.						n.s.
C x Jul 01	0.15	0.14																
C x Oct 01	-0.17	0.14																
C x Feb 02	-0.28	0.14																
F x Jul 01	-0.04	0.14																
F x Oct 01	-0.13	0.14																
F x Feb 02	-0.31	0.14																

Table 1.- Summary of generalized linear models testing the main factors treatment (three levels), date (four levels) and their interaction on community traits: taxonomic richness, diversity (weighted Simpson Index, transformed as Dw^2) and total dry mass (g, $\log_{10}$ transformed). When the interaction was not significant ($P > 0.05$) it was removed from the analysis. Pond and plot were included as random factors using the GLIMMIX procedure with an identity link function and normal error distribution. See methods for details. All-bird exclosure treatment and date Jun 02, and all combinations with at least one of these levels, were aliased. C = controls, F = flamingo exclosures.

	FACTOR	Total weight		Diversity	
		T_{159}	p	T_{165}	p
Treatment	C vs F	-4.04	0.0002	-0.99	0.6
	C vs A	-6.86	<0.0001	-2.93	0.01
	F vs A	-2.82	0.02	-1.93	0.13
Date	Jul 01 vs Oct 01	1.69	0.3	1.01	0.8
	Jul 01 vs Feb 02	2.10	0.16	0.1	1.0
	Jul 01 vs Jun 02	-0.53	0.9	-3.29	0.007
	Oct 01 vs Feb 02	0.44	0.9	-0.96	0.8
	Oct 01 vs Jun 02	-2.31	0.1	-4.51	<0.0001
	Feb 02 vs Jun 02	-2.73	0.04	-3.58	0.003

Table 2.- Post-hoc least-squared means tests comparing pairwise differences among levels of the main effects treatment (three levels) and date (four levels) that were significant when analyzingdiversity (weighted Simpson Index) and total dry mass. Post-hoc tests were not performed when main effects were not significant (see Table 1). P-values were adjusted with Tukey tests. C = controls, F = flamingo exclosures, A = all-bird exclosures.

FACTOR	Other Polychaetes						*P. antipodarum*						Chironomids						*C. torosa*					
	Estimate	SE	df_N	df_D	F	P	Estimate	SE	df_N	df_D	F	P	Estimate	SE	df_N	df_D	F	p	Estimate	SE	df_N	df_D	F	p
Intercept	5.82	0.28					1.14	0.62					4.4	0.18					6.19	0.2				
Treatment (T)			2, 165		8.58	0.0003			2, 165		6.99	0.001			2, 165		17.12	<0.0001			2, 165		1.48	0.2
C	-0.70	0.18					-0.51	0.18					-0.79	0.14					-0.2	0.11				
F	-0.17	0.18					-0.62	0.18					-0.43	0.13					-0.09	0.11				
Date (D)			3, 165		1.05	0.4			3, 165		33.21	<0.0001			3, 165		30.53	<0.0001			3, 165		36.26	<0.0001
Jul 01	-0.013	0.23					1.73	0.22					-1.05	0.17					-1.08	0.15				
Oct 01	0.32	0.21					0.17	0.22					0.41	0.16					-1.38	0.14				
Feb 02	0.13	0.21					-0.17	0.22					-0.59	0.16					-0.92	0.14				
TxD						n.s.						n.s.						n.s.						n.s.

FACTOR	*N. diversicolor*					
	Estimate	SE	df_N	df_D	F	P
Intercept	-1.99	0.89				
Treatment (T)			2, 165		1.79	0.2
C	-0.97	0.52				
F	-0.52	0.49				
Date (D)			3, 165		4.27	0.006
Jul 01	1.89	0.64				
Oct 01	0.98	0.62				
Feb 02	-0.0.3	0.67				
TxD						n.s.

Table 3.- Summary of generalized linear models testing the main effects treatment (three levels) and date (four levels) and their interaction on invertebrate counts. Pond and plot were included as random factors using the GLIMMIX procedure with a log link function and negative binomial error distribution. See methods for details. All-bird exclosure treatment and date Jun 02 were aliased. C = controls, F = flamingo exclosures.

		Other Polychaetes		*P. antipodarum*		Chironomids		*C. torosa*		*N. diversicolor*	
	FACTOR	**T_{165}**	**P**	**T_{165}**	**p**	**T_{165}**	**p**	**T_{165}**	**p**	**T_{165}**	**p**
Treatment	C vs F	-3.01	0.009	0.58	0.8	-2.64	0.02				
	C vs A	-3.97	0.0003	-2.89	0.01	-5.84	<0.0001				
	F vs A	-0.96	0.6	-3.47	0.002	-3.21	0.005				
Date	Jul 01 vs Oct 01			7.43	<0.0001	-8.79	<0.0001	2.06	0.2	1.63	0.4
	Jul 01 vs Feb 02			8.89	<0.0001	-2.75	0.03	-1.12	0.7	3.07	0.01
	Jul 01 vs Jun 02			7.70	<0.0001	-6.13	<0.0001	-7.30	<0.0001	2.95	0.02
	Oct 01 vs Feb 02			1.60	0.4	6.48	<0.0001	-3.44	0.004	1.63	0.4
	Oct 01 vs Jun 02			0.76	0.9	2.59	0.05	-10.05	<0.0001	1.55	0.4
	Feb 02 vs Jun 02			-0.75	0.9	-3.70	0.002	-6.74	<0.0001	-0.05	1.0

Table 4**.-** Post-hoc least-squared means tests comparing pairwise differences among levels of the main effects treatment (three levels) and date (four levels) that were significant in analyses of invertebrate counts (see Table 3). Post-hoc tests were not performed when main effects were not significant. P-values were adjusted with Tukey tests. C = controls, F = flamingo exclosures, A = all-bird exclosures.

	Other Polychaetes (w)						*P. antipodarum* (w)						Chironomids(w)						*C. torosa*(w)					
FACTOR	**Estimate**	**SE**	**df_N**	**df_D**	**F**	**p**	**Estimate**	**SE**	**df_N**	**df_D**	**F**	**p**	**Estimate**	**SE**	**df_N**	**df_D**	**F**	**p**	**Estimate**	**SE**	**df_N**	**df_D**	**F**	**p**
Intercept	-4.48	0.07					-2.94	0.1					-3.95	0.07					-4.14	0.05				
Treatment (T)			2, 158		3.03	0.05			2, 132		14.37	<0.0001			2, 162		0.46	0.6			2, 159		0.05	0.9
C	-0.14	0.06					-0.42	0.08					0.04	0.06					0.01	0.03				
F	-0.08	0.06					-0.16	0.08					0.05	0.06					0.01	0.03				
Date (D)			3, 158		5.10	0.002			3, 132		4.75	0.004			3, 162		21.99	<0.0001			3, 159		5.49	0.001
Jul 01	-0.01	0.07					-0.13	0.1					0.23	0.07					-0.15	0.04				
Oct 01	0.04	0.07					-0.24	0.09					-0.03	0.07					-0.06	0.04				
Feb 02	-0.19	0.07					-0.34	0.1					0.46	0.07					-0.11	0.04				
TxD						n.s.						n.s.						n.s.						n.s.

	L. hookeri (w)						*C. orientale* (w)					
FACTOR	**Estimate**	**SE**	**df_N**	**df_D**	**F**	**p**	**Estimate**	**SE**	**df_N**	**df_D**	**F**	**p**
Intercept	-3.3	0.15					-3.83	0.12				
Treatment (T)			2, 48		0.52	0.6			2, 70		1.24	0.3
C	-0.17	0.17					-0.15	0.1				
F	-0.05	0.14					-0.10	0.09				
Date (D)			3, 48		4.75	0.006			3, 70		4.10	0.001
Jul 01	0.08	0.16					-0.05	0.13				
Oct 01	-0.42	0.22					-0.25	0.12				
Feb 02	-0.73	0.3					0.14	0.11				
TxD						n.s.						n.s.

Table 5.- Summary of generalized linear models testing the main effects treatment (three levels) and date (four levels) and their interaction on invertebrate individual mean dry mass (g, $\log_{10}$ transformed). When the interaction was not significant ($P > 0.05$) it was removed from the analysis. Pond and plot were included as random factors using the GLIMMIX procedure with an identity link function and normal error distribution. See methods for details. All-bird exclosure treatment and date Jun 02 were aliased. C = controls, F = flamingo exclosures.

	FACTOR	Other Polychaetes (w)		*P. antipodarum* (w)		Chironomids (w)		*C. torosa* (w)		*L. hookeri* (w)		*C. orientale* (w)	
		T_{158}	p	T_{132}	p	T_{162}	p	T_{159}	p	T_{48}	p	T_{70}	P
Treatment	C vs F	-0.94	0.6	-3.27	0.004								
	C vs A	-2.44	0.04	-5.32	<0.0001								
	F vs A	-1.50	0. 3	-2.01	0.1								
Date	Jul 01 vs Oct 01	-0.85	0.8	1.21	0. 6	3.37	0.003	-2.30	0.1	2.57	0.06	1.46	0.5
	Jul 01 vs Feb 02	2.58	0.05	2.26	0.1	-3.27	0.007	-0.85	0.8	3.05	0.02	-1.50	0.4
	Jul 01 vs Jun 02	-0.20	0.9	-1.38	0.5	3.09	0.01	-3.77	0.001	0.49	0.9	-0.41	0.9
	Oct 01 vs Feb 02	3.64	0.002	1.13	0.7	-7.19	<0.0001	1.48	0.5	1.06	0.7	-3.48	0.005
	Oct 01 vs Jun 02	0.64	0.9	-2.58	0.05	-0.42	0.9	-1.70	0.3	-1.94	0.2	-2.16	0.1
	Feb 02 vs Jun 02	-2.82	0.03	-3.57	0.003	6.61	<0.0001	-3.04	0.01	-2.70	0.05	1.31	0.6

Table 6.- **.-** Post-hoc least-squared means tests comparing pairwise differences among levels of the main effects treatment (three levels) and date (four levels) when these were significant in analyses of invertebrate individual mean mass (see Table 5). Post-hoc tests were not performed when main effects were not significant. P-values were adjusted with Tukey tests. C = controls, F = flamingo exclosures, A = all-bird exclosures.

		Jul 01		Oct 01		Feb 02		Jun 02	
		t	p	t	p	t	p	t	p
Total weight df=159	C vs. F	-0.55	1.0	-2.82	0.2	-2.23	0.5	-2.47	0.4
	C vs. A	-1.10	0.9	-4.40	0.001	-5.57	<.0001	-2.65	0.3
	F vs. A	-0.55	1.0	-1.57	0.9	-3.34	0.05	-0.18	1.0

Table 7.- Post-hoc least-squared means tests on the significant interaction for the model of total dry mass (see Table 1) among treatment and date, comparing pairwise differences among treatments (factor of three levels) for different dates (factor of four levels). P-values were adjusted with Tukey tests. C = controls, F = flamingo exclosures, A = all-bird exclosures.

Class	Groups identified	Apr 01		Jul 01		Jul 01		Oct01		Nov01		Feb 02		Mar 02		Jun 02	
		N	B (g/m^2)	N	B (g/m^2)	N	B (g/m^2)	N	B (g/m^2)	N	B (g/m^2)	N	B (g/m^2)	N	B (g/m^2)	N	B (g/m^2)
Polychaeta	O. Phyllodocida F. Nereidae *Nereis diversicolor* (Müller 1776)	149	*0.361*	162	*0.431*	75	*0.458*	52	*0.117*	6	*0.015*	150	*0.065*	357	*0.050*	74	*0.836*
	O. Terebellidei F. Ampharetidae *Alkmaria romijni* (Horst 1919) O. Canalipalpata F. Spionidae *Streblospio shrubsolli* (Buchanan 1890)	17977	*1.327*	16882	*1.465*	13709	*1.291*	26901	*2.914*	7143	*0.484*	19265	*1.678*	10737	*0.613*	13940	*1.421*
Ostracoda	O. Podocopa F. Cytherideidae *Cyprideis torosa* (Jones 1857)	8527	*1.218*	10011	*2.229*	9137	*1.410*	4679	*0.804*	3965	*0.607*	7538	*1.312*	7213	*1.306*	19207	*3.955*
Gastropoda	O. Neotaeniglossa F. Hydrobiidae *Potamopyrgus antipodarum* (Gray 1843)	195	*0.171*	1640	*3.515*	1306	*2.614*	502	*1.170*	205	*0.173*	200	*0.295*	150	*0.192*	293	*0.803*
Malacostraca	O. Isopoda F. Sphaeromatidae *Lekanesphaera hookeri* (Leach 1814)	16	*0.056*	427	*0.860*	447	*0.802*	40	*0.052*	1	*0*	15	*0.017*	3	*0*	92	*0.324*
	F. Anthuridae *Cyathura carinata* (Kröyer 1848)	0	*0*	14	*0.021*	24	*0.032*	24	*0.043*	0	*0*	2	*0.003*	0	*0*	10	*0.001*
	O. Amphipoda F. Corophiidae *Corophium orientale* (Schllenberg 1928)	1327	*0.556*	325	*0.148*	511	*0.236*	505	*0.150*	257	*0.067*	1050	*1.313*	306	*0.177*	1566	*0.961*
	O. Decapoda F. Palaemonidae *Palaemonetes varians* (Leach 1814)	0	*0*	0	*0*	1	*0.041*	1	*0.005*	0	*0*	0	*0*	0	*0*	0	*0*
Euentomata	O. Coleoptera F. Hydrophilidae *Berosus sp.* *Enochrus sp.*	0	*0*	2	*0*	3	*0.022*	0	*0*	0	*0*	0	*0*	0	*0*	0	*0*
	O. Hemiptera F. Corixidae *Sigara stagnalis* (Leach 1817)	86	*0.055*	5	*0.003*	0	*0*	0	*0*	0	*0*	0	*0*	0	*0*	6	*0.006*
	O. Odanata F. Coenigrionidae *Ischnura graellsi* (Rambur 1842)	0	*0*	0	*0*	1	*0.015*	0	*0*	0	*0*	0	*0*	0	*0*	0	*0*
	O. Diptera F. Chironomidae Tr. Chironomini	1375	*0.331*	880	*0.531*	1013	*0.464*	4833	*2.492*	2804	*0.963*	1776	*2.254*	868	*1.109*	3148	*1.491*
	F. Ephydridae *Ephydra sp.*	0	*0*	1	*0.001*	1	*0.009*	0	*0*	0	*0*	0	*0*	0	*0*	0	*0*
Turbelleria	O. Catenulida	943	*0.009*	12	*0.001*	12	*0.001*	0	*0*	35	*0.002*	13	*0.001*	262	*0.006*	55	*0.002*

Table 8.- Total number (N) of invertebrates and dry mass (B) of each taxonomic group analysed. All samples within each period were pooled together regardless the treatment. Identification was made with the following keys; Tachet et al (2003), Jansson (1986), Castello (1986), Askew 1988, Ruffo (1989), Ortiz and Jimeno (2001), Argano (1979), Ghetti and Mc Kenzie (1981), Fauvel (1923, 1927), Holthe (1986). Some identifications were done with the help of specialists (see acknowledgements) and confirmed in Baldó et al (2001) for the same estuary. In the case of polychaetes and Coleoptera, it was not practical to identify all individuals to a species level.

Fig 1.- Aerial photograph of craters made by Greater flamingos in the ponds in Veta la Palma. The flamingos were flushed immediately before the photo was taken. Author Héctor Garrido/Equipo de Seguimiento de Procesos Naturales.

Fig 2.- Map of the study area showing the location within the Iberian peninsula. Ponds used in the study are labelled with their local name. A schematic drawing shows an example of the position of experimental blocks and of the different treatments within each block. Source: Digital Orthophoto of Andalusia (1998-1999). Projection UTM Datum European 1950, Spain and Portugal Zone 30.

Fig 3.- The upper plot shows the aerial counts of waterbirds in Veta la Palma from March 2001 to June 2002. Totals represent all birds counted in the area, including shorebirds, gulls, etc. The lower plot shows the biomass (kg/ha) of flamingos and waterfowl during experimental periods in Veta la Palma. Calculations were based on body masses in Hoyo et al (1992).

Fig 4.- Geometric mean and back-transformed SE of taxa richness and total dry mass, and arithmetic mean and SE of diversity index at the end of each experimental period, including all data.

Fig 5.- Geometric mean and back-transformed SE of counts for polychaetes (excluding nereids), *Nereis diversicolor*, *Potamopyrgus antipodarum*, chironomid larvae and *Cyprideis torosa* at the end of each experimental period.

Fig 6.- Geometric mean and back-transformed SE of mean individual dry mass (in g) for polychaetes (excluding nereids), *Potamopyrgus antipodarum*, chironomid larvae, *Cyprideis torosa*, *Lekanesphaera hookeri* and *Corophium orientale*.

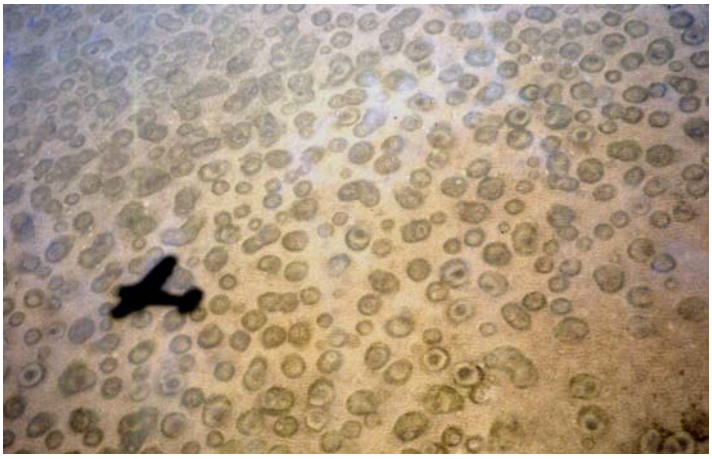


Fig 1.-

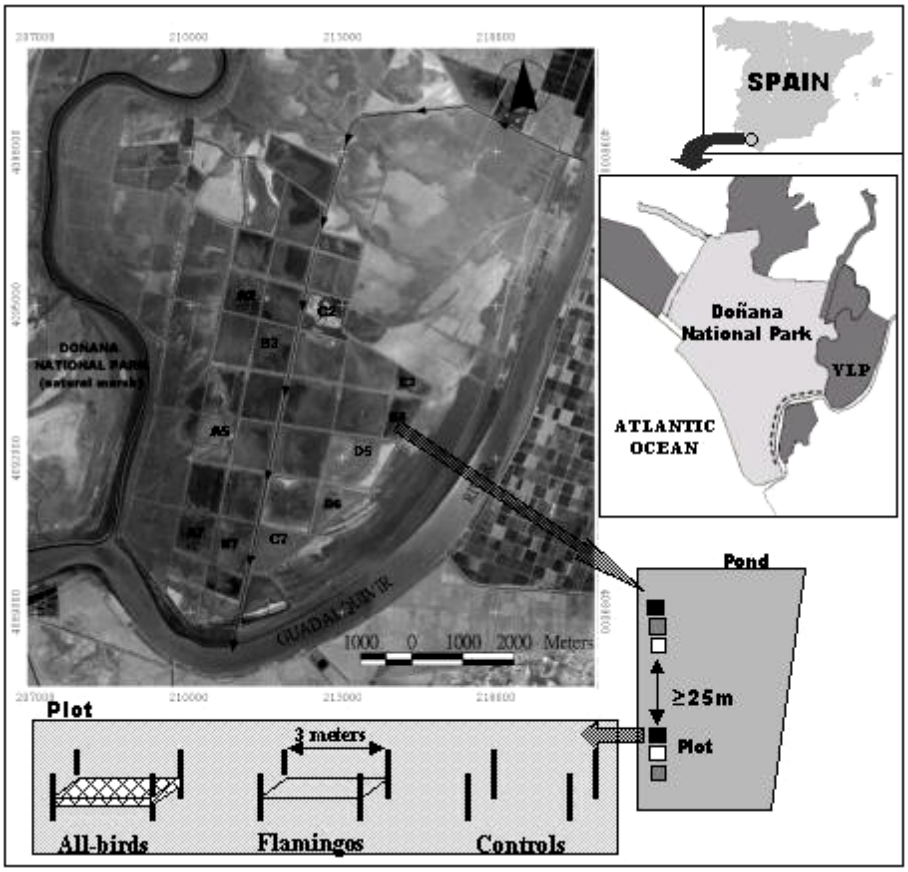

SPAIN
Doñana
National Park
VLP
ATLANTIC
OCEAN
DOÑANA
NATIONAL PARK
(natural mark)
GUADALQUIVIR
1000 0 1000 2000 Meters
Pond
≥25m
Plot
Plot
3 meters
All-birds
Flamingos
Controls

Fig.-2

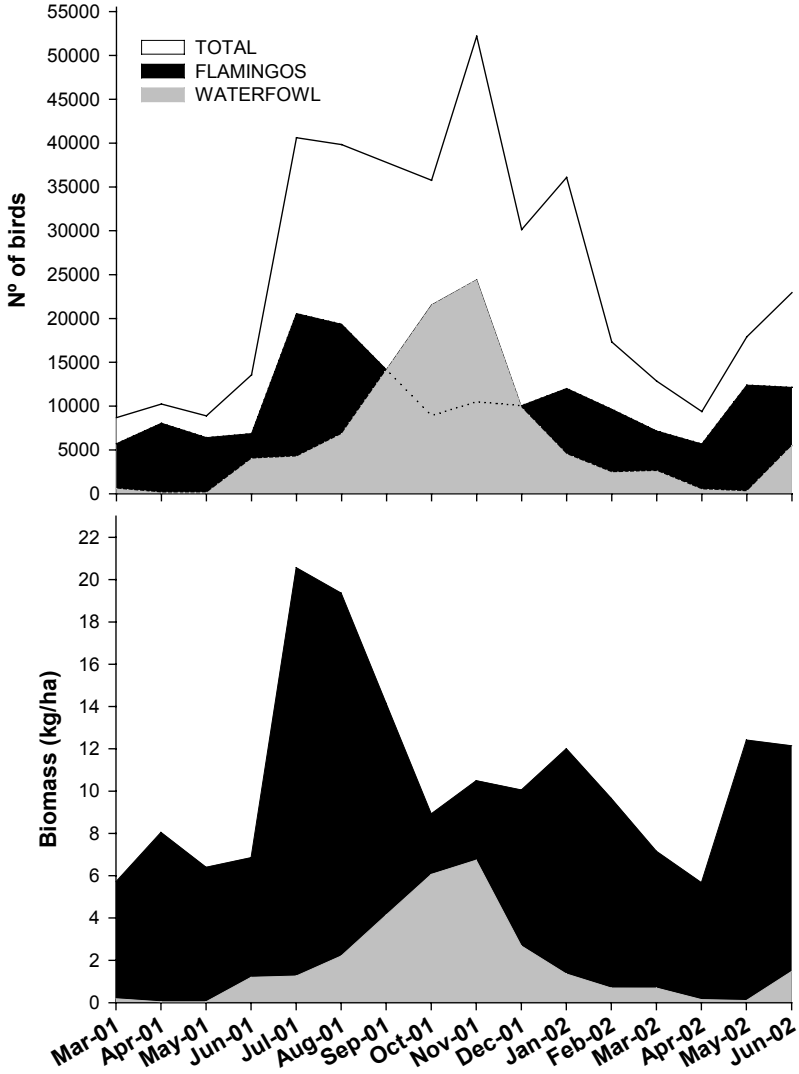

TOTAL
FLAMINGOS
WATERFOWL
Nº of birds
55000
50000
45000
40000
35000
30000
25000
20000
15000
10000
5000
0
Biomass (kg/ha)
22
20
18
16
14
12
10
8
6
4
2
0
Mar-01
Apr-01
May-01
Jun-01
Jul-01
Aug-01
Sep-01
Oct-01
Nov-01
Dec-01
Jan-02
Feb-02
Mar-02
Apr-02
May-02
Jun-02

Fig 3.-

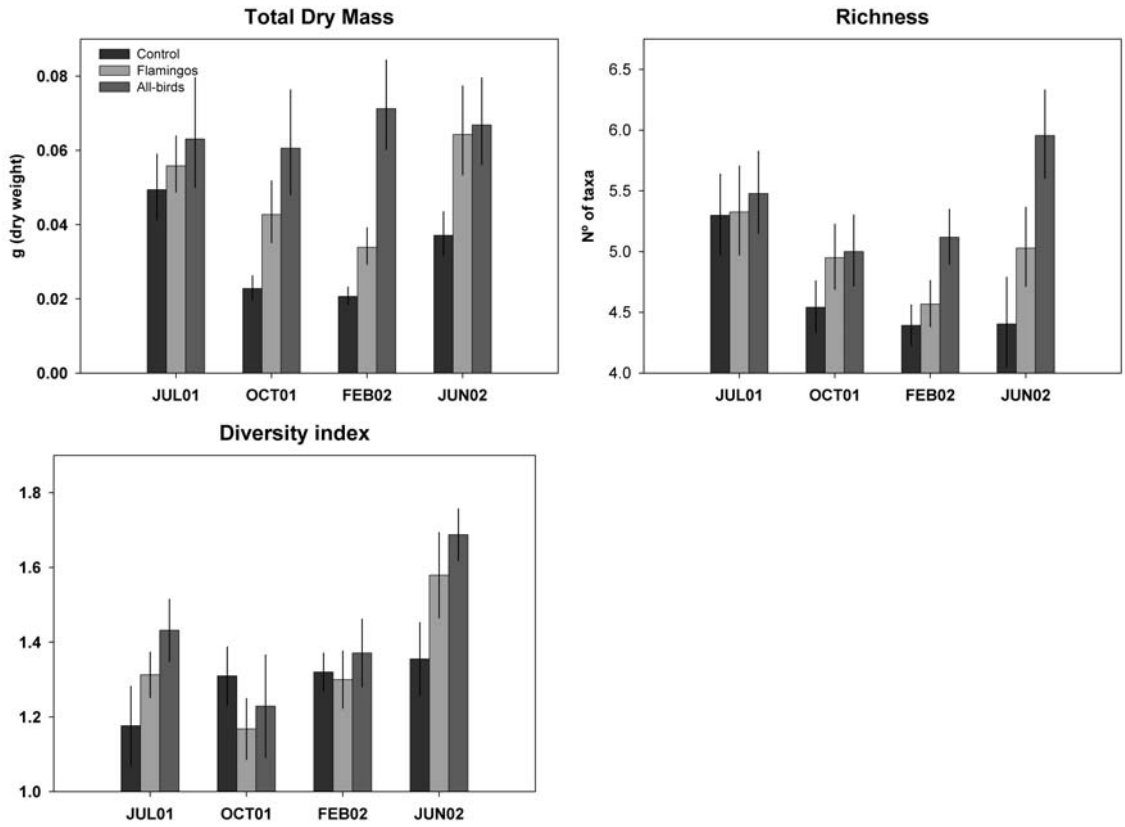

Total Dry Mass
Control
Flamingos
All-birds
g (dry weight)
0.00
0.02
0.04
0.06
0.08
JUL01
OCT01
FEB02
JUN02
Richness
Nº of taxa
4.0
4.5
5.0
5.5
6.0
6.5
JUL01
OCT01
FEB02
JUN02
Diversity index
1.0
1.2
1.4
1.6
1.8
JUL01
OCT01
FEB02
JUN02

Fig 4.-

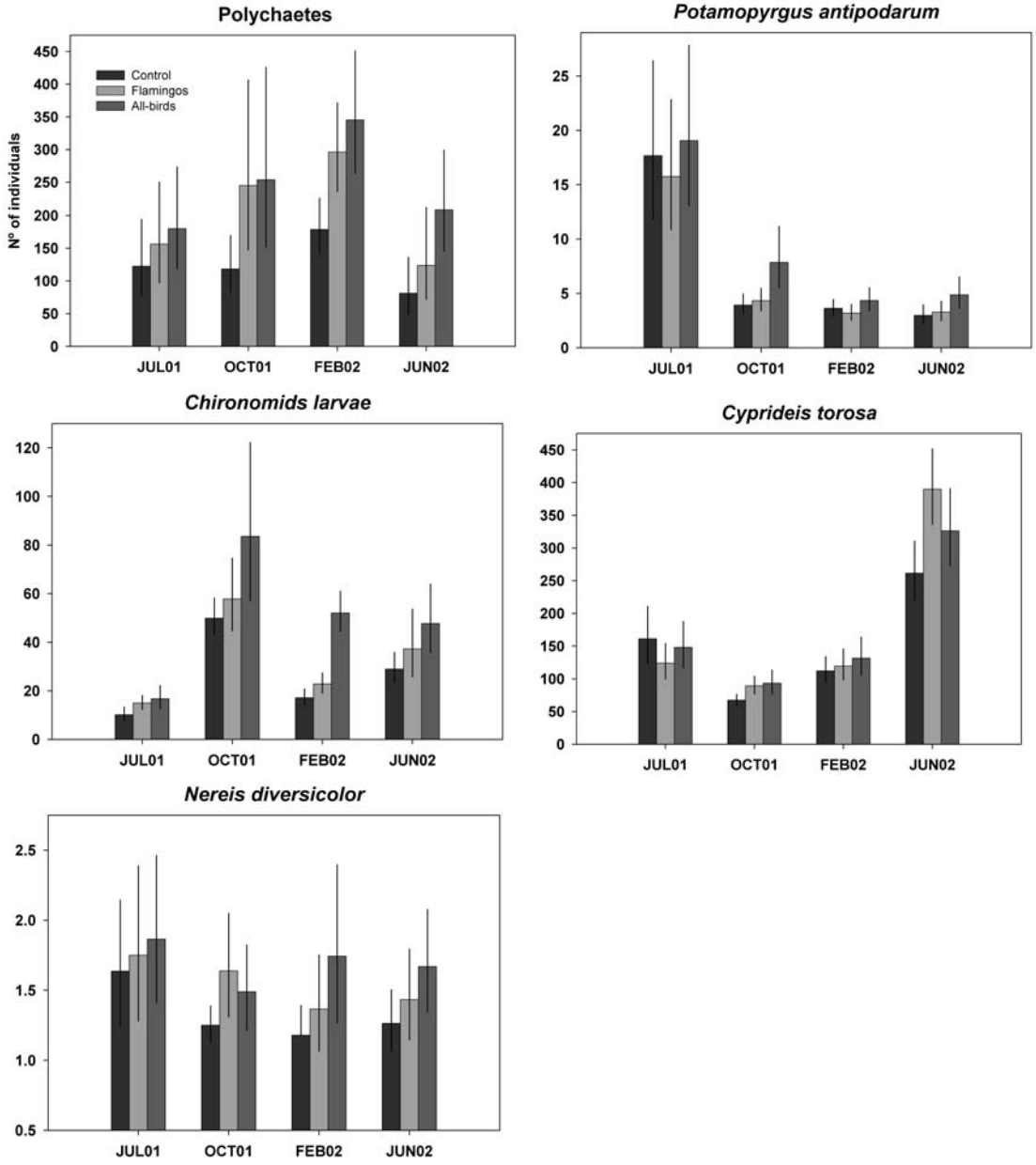

Polychaetes
Nº of individuals
Control
Flamingos
All-birds
JUL01
OCT01
FEB02
JUN02
Potamopyrgus antipodarum
Chironomids larvae
Cyprideis torosa
Nereis diversicolor

Fig 5.-

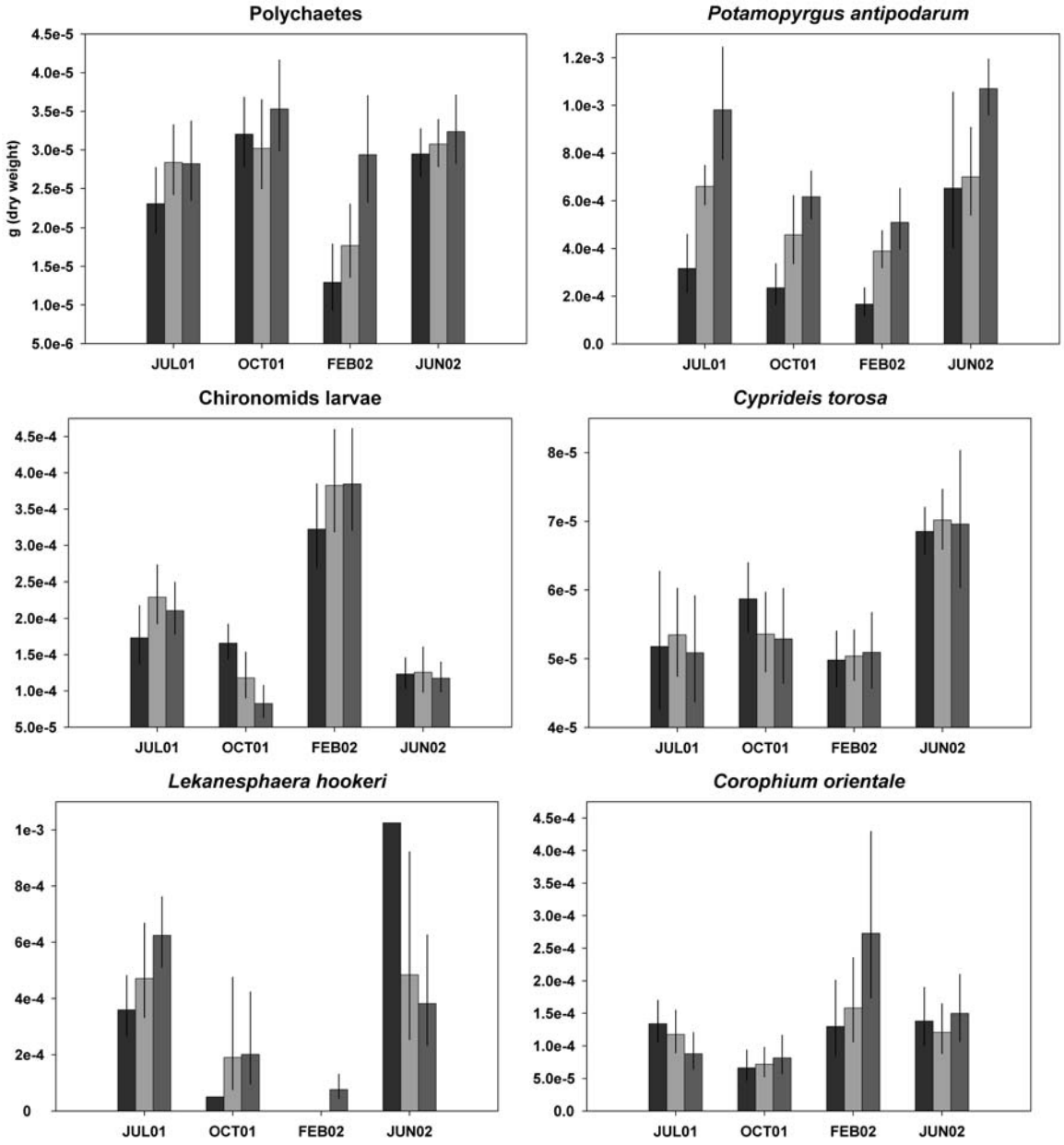

Polychaetes
Potamopyrgus antipodarum
Chironomids larvae
Cyprideis torosa
Lekanesphaera hookeri
Corophium orientale
g (dry weight)
JUL01
OCT01
FEB02
JUN02

Fig 6.-

CAPÍTULO 3.

Efecto de las aves acuáticas sobre los invertebrados de la columna de agua / Waterbirds effect on water column invertebrates.

Basado en el manuscrito original: Rodríguez-Pérez, H., and Green, A.J. Waterbirds have strong influence on invertebrates in the water column in shallow lakes. A case study in south-west Spain. .

Resumen

Se ha prestado poca atención al papel que tienen las aves acuáticas estructurando las comunidades de invertebrados acuáticos, especialmente para el caso de los invertebrados no bentónicos. Hemos usado distintos tipos de cercados de exclusión para excluir flamencos o todos el conjunto de aves acuáticas (flamencos, patos y fochas) en exclusiones de 3x3 metros, y las hemos comparado con controles, en 11 balsas de piscicultura extensiva construidas en una antigua marisma, en Doñana,. Se realizaron cuatro experimentos de tres meses de duración en diferentes momentos de un ciclo anual, considerando las diferentes densidades de aves. La diversidad, la riqueza taxonómica y la biomasa total de los invertebrados presentes en la columna de agua fueron significativamente superiores en los cercados que excluían a todas las aves, que en los controles y los cercados de flamencos. La biomasa media en cada periodo fue de entre el 74 y el 94% más alta para los exclusiones completas que para los controles. No hubo interacción significativa Tratamiento x Periodo para estos descriptores de la comunidad, indicando que las aves tuvieron efectos importantes a lo largo de todo el año. A nivel de especie, el número de camarones (*Palaemonetes varians*), anfípodos (*Corophium orientale*), isópodos (*Lekanespahera hookeri*), gasterópodos (*Potamopyrgus antipodarum*), y de coríxidos (*Sigara stagnalis*) fueron significativamente superiores en las exclusiones completas que en los controles. Los isópodos fueron también más abundantes en los cercados de flamencos que en los controles. Sin embargo, el número de ostrácodos (*Cyprideis torosa*) fue significativamente menor en los cercados que excluían a todas las aves. La masa media individual de los isópodos y los camarones fue significativamente superior en las exclusiones totales que en los controles, y la masa media de las larvas de quironómidos fue mayor en las exclusiones de flamencos que en los controles. Nuestro resultado pone de manifiesto que las aves acuáticas tienen una gran influencia sobre las comunidades y las poblaciones de invertebrados no bentónicos. Las aves consumen macrófitos al igual que invertebrados, y la influencia relativa de dos consumos sigue sin estar clara en los efectos de las aves acuáticas sobre los invertebrados no bentónicos. Las densidades registradas de aves acuáticas en nuestra área de estudio fueron relativamente bajas, en comparación otros estudios de los efectos de las ave acuáticas en ecosistemas acuáticos, y nuestros resultados sugieren que la importancia de las aves acuáticas estructurando este tipo de ecosistemas ha sido infravalorada.

Palabras clave: Cercados de exclusión, aves acuáticas, flamencos, depredación, necton, zooplancton, invertebrados epifíticos, lagos someros.

Abstract

Little attention has been paid to the role of waterbirds in structuring communities of aquatic invertebrates, and this is especially true for non-benthic invertebrates. In 11 extensive fish ponds constructed on a former marshland in Doñana (SW Spain), we used separate exclosure designs to exclude flamingos or all waterbirds (flamingos, ducks and coot) from 3 x 3 m plots and compare them with control plots. Four experiments were conducted for three month periods at different points of the annual cycle with varying bird densities. The diversity, taxa richness and total biomass of invertebrates present in the water column were significantly higher in all-bird exclosures than in controls or flamingo exclosures. Mean biomass for each period was 74 to 94% higher in all-bird exclosures than in controls . There were no significant treatment x season interactions for these community parameters, indicating that birds had important effects throughout the year. On a taxon level, numbers of the shrimp *Palaemonetes varians*, the amphipod *Corophium orientale*, the isopod *Lekanesphaera hookeri*, the gastropod *Potamopyrgus antipodarum* and the corixid *Sigara stagnalis* were all significantly higher in all-bird exclosures than in controls. Isopods were also more abundant in flamingo exclosures than controls. In contrast, numbers of the ostracod C*yprideis torosa* were significantly lower in all-bird exclosures. The mean mass of individual shrimps or isopods was significantly higher in all-bird exclosures than in controls, and the mean mass of chironomid larvae was higher in flamingo exclosures than controls. Our results make it clear that waterbirds can have a major influence on non-benthic invertebrate populations and communities. As well as predating on the invertebrates, waterbirds consumed submerged plants and disturbed sediments, and the relative influence of these processes remains unclear. The densities of waterbirds in our study were relatively low compared with other studies of their effect on aquatic ecosystems, and our findings suggest that the importance of waterbirds in structuring such ecosystems has previously been overlooked.

Keywords: Exclosures, waterfowl, flamingos, predation, nekton, zooplankton, epiphytic invertebrates, shallow lakes.

INTRODUCTION

There is a wealth of literature about waterbirds, but relatively little experimental work addressing their influence on aquatic ecosystems. Previous research has focussed on the use of exclosure experiments to investigate the role of waterbirds in herbivory of aquatic plants (see Lodge et al. 1998, Marklund et al. 2002 for reviews) and in predation of benthic invertebrates, particularly annelids, chironomids and bivalves (Botto et al. 1998, Marklund & Sandsten 2002, Hamilton & Nudds 2003). However, there has been almost no attention paid to the effect of birds on the abundance of invertebrates found in the water column (but see Cooper 1984), despite the fact that such invertebrates are important prey items for ducks, flamingos and other waterbirds (Vareschi and Jacobs 1984, Rubega and Inouye 1994, deSzalay and Resh 1997, Kear 2005). In contrast, it is well known that fish predation has profound effects on non-benthic invertebrate communities (Gilinsky 1984, Richardson et al. 1990, Rubega and Inouye 1994, Diehl 1995, Crowder et al 1998, Wetzel 2001).

Here we present an exclosure study comparing the effects of Greater Flamingos (*Phoenicopterus ruber*) and waterfowl (ducks and coots) on invertebrates in the water column in shallow brackish marshes in Doñana, south-west Spain, during distinct three months periods of the annual cycle. Both groups of birds have additive effects on submerged macrophytes and benthic infauna in our study area (Rodríguez-Pérez and Green 2006, unpublished manuscript). In this study, we consider the effects of these two waterbird groups on the abundance and size of different invertebrates in the water column, as well as on taxonomic richness, diversity and total biomass. As well as direct effects from predation, these invertebrates may also be influenced by the reductions in abundance of vegetation caused by the waterbirds (Crowder et al 1998). Flamingos disturb sediments (Glassom & Branch, 1997a,b, Rodríguez-Pérez & Green 2006), and this bioturbation might also influence the distribution of invertebrates in the water column.

Ours is a novel study comparing the effects of waterbirds on invertebrates in the water column at different times of the annual cycle. As far as we know, ours is the first study of non-benthic invertebrates to compare the impacts of different groups of waterbirds, or to assess the influence of changes in waterbird density during the annual cycle.

Study area

We carried out exclosure experiments in Veta la Palma (36^{o}57'N, 6 ^{o}14'W), a private fish-farm divided into 52 ponds within Doñana Natural Park. The ponds were constructed in

1992-1993 on top of what was natural marshland in the Guadalquivir estuary (Fig. 2). All the ponds are shallow (average 30 cm, maximum depth 50 cm) and flat-bottomed with a total combined surface area of 2997 ha. The fish species cultured are European Seabass (*Dicentrarchus labrax*), Flathead Mullet (*Mugil cephalus*), Gilthead seabream (*Sparus auratus*) and eels (*Anguilla anguilla*) as well as Atlantic ditch shrimp (*Palaemonetes varians*). The alien killifish *Fundulus heteroclitus* is also abundant, as is the exotic crab *Rhithropanopeus harrisii*. Each pond is dried out under rotation approximately every two years to extract fish. Ponds are interconnected via canals, and permanent flow of water taken from the Guadalquivir estuary maintains dissolved oxygen levels (see Frisch et al. 2005, Rodríguez-Pérez & Green 2006 for more details).

The dominant submerged macrophyte is *Ruppia maritima*, forming extensive beds during spring and early summer. Small patches of *Potamogeton pectinatus* occur in some ponds in years of relatively low salinity. Most of the shoreline is bare mud, and regular dredging to prevent siltation of peripheral canals used to extract fish from the ponds has restricted development of vegetation. Salinity during our study varied from 7 g/l during winter months of high rainfall to 15 g/l at the end of September, after the dry summer months typical of the Mediterranean region. pH ranged from 9.3 to 10.4.

MATERIALS AND METHODS.

Aerial counts of waterbirds in Veta la Palma were carried out monthly during the study, during surveys of a wider area including Doñana National Park and its surroundings. All waterbirds were counted, including those on the water and those in flight (most of which were flushed by the plane).

Eight ponds were used in each 3 month experimental period. Three ponds were substituted in the following periods when they were drained in order to extract fish, making a total of 11 different ponds used during the study (range 26.1-114.4 ha, making a total of 696.84 ha). All exclosures and controls were 3 x 3m in size, and we used two exclosure treatments. One treatment used a 2 cm mesh nylon net tied to a square PVC frame suspended from four iron poles (2 m long, 10 mm diameter) pushed into the pond bottom in each corner. Wires were also attached from the top of the poles to the centre of the net to prevent it from touching the water surface. At the sides of the square frame, 20 cm of net hung vertically down to prevent birds from passing underneath. This design allowed fish to enter but decreased utilisation by ducks, coot and other birds. The netting did not affect water movement. There was no algal growth and shading effects were close to zero. The second treatment excluded only flamingos by taking advantage of their much greater height. Four iron poles were placed in each corner of the square as before, and a wire tied around

them at a height of 70-75 cm from the bottom (i.e. 30-65 cm above the water surface). This height was selected after trials confirming that other birds pass comfortably underneath the wire and freely enter the exclosures. The controls consisted merely of the four iron poles. Eurasian coots *Fulica atra* and ducks were frequently observed feeding inside controls and flamingo exclosures, whilst no birds were ever observed inside all-birds exclosures. It is also possible that Black-tailed Godwit (*Limosa limosa*) and other larger waders occasionally entered the shallowest plots.

Each experimental block contained one exclosure of each kind plus a control, separated by approximately 10 m. Treatments were randomly located within each block. There were two experimental blocks in each pond, separated by approximately 25 m. Both blocks were placed in the western part of each pond, aligning all exclosures and controls at an equal distance from the pond edge (approximately 20 m for the first experiment), so as to expose them equally to the dominant southwesterly wind with an equal and relatively low fetch. Invertebrates in the water column were sampled twice in each exclosure and control plot, immediately after establishing the plots, and three months after installing them. At the same time, benthic invertebrates in the sediments and submerged macrophytes were also sampled using different methods (Rodríguez-Pérez and Green 2006, authors unpublished data). The experiment was repeated four times between April 2001 and June 2002, moving the exclosures and controls each time approximately 10 m in towards the centre of the pond, but preserving the layout described above.

The first experimental period ran from 3 April to 11 July 2001 (Jul 01 from hereon) coinciding roughly with the breeding period for waterbirds. The second period, from 13 July to 30 October 2001 (Oct 01 from hereon), corresponded with the post-breeding period. The third period from 22 November 2001 to 27 February 2002 (Feb 02 from hereon) covered the wintering period. The fourth period from 7 March to 6 June 2002 (Jun 02 from hereon) covered the pre-breeding and early breeding periods. Each individual exclosure or control was set for 70-93 days in Jul 01, 82-92 days in Oct 01, 95-105 days in Feb 02 and 88-96 days in Jun 02. In a given pond and period, all exclosures and controls were established or sampled on the same day. There was a slight overlap between experimental periods caused by the time required to sample and move exclosures and controls in the ponds. This usually took two weeks, but bad weather caused some delays.

When plots were established, two samples of invertebrates were taken from the inside edge, so as to avoid stepping inside. Three months later, two samples were taken from at least one meter inside the edge to avoid edge effects. A PVC pipe section of 20 cm diameter was inserted into the mud to isolate the water within. Using a plastic jar, all the water was then scooped out and sieved through a 250 μm mesh, taking care not to extract sediments. The sieved material was then fixed with formalin. The water depth at sampling points ranged

from 10 to 39 cm (mean+s.e. = 27±8 cm), within the range accessible to dabbling ducks when feeding (Pöysä 1983). Samples were later placed under a microscope and the animals sorted into taxa, counted and separated and stored in formalin. We identified invertebrates to species level whenever practical. Prior to statistical analysis, we summed data from the two samples taken from each plot because they were not spatially independent.

To calculate dry mass, each taxon sample was dried for 24 hour at 80ºC and then weighed to the nearest 0.0001 g. When samples weighed less than the balance precision, a mass of 0.00005 g was assumed. Individual mean dry mass was assessed for each sample by dividing the dry mass by the number of individuals. Owing to their small size, it was not possible to weigh individual samples of copepods and ostracods in a satisfactory manner. An overall mean dry mass for copepods was calculated by weighing 15 samples with large numbers (between 423 and 1085) of individuals. For the ostracod *Cyprideis torosa*, the mean weight was calculated from benthic samples (authors, unpublished manuscript). Using these mean weights and the number of individuals, the mass of ostracods and copepods was estimated when calculating the total sample dry mass.

Statistical methods

We analyzed the effects of different treatments (all-birds exclosures, flamingo exclosures, controls), experimental period, depth and the treatment x period interaction on the non-benthic invertebrate community using Generalized mixed Linear Models (GLMs, McCullagh and Nelder 1989). Interactions were only retained in the models when they were significant. Dependent variables were counts (including zeros) and mean individual mass (as a body size estimate) of different taxa, as well as total sample dry mass, richness (number of taxa per sample) and a diversity index. We used a modified Simpson index (Dw) because it reflects the importance of both diversity and abundance (Sherfy and Kirkpatrick 2003). In an unmodified Simpson index, dominance has most weight, whereas taxa richness has the most weight in the Shannon index (Magurran 1988). Taxa richness and diversity index were calculated based on the finest taxonomical level reached, mixing species level with broader levels of identification.

Pond and experimental block nested within ponds were included as random factors using the GLIMMIX procedure (SAS Institute 2005). Treatment and sampling period were included as fixed factors of three and four levels respectively. *Post-hoc* analyses of least-squared means and Tukey-adjusted P values identified significant differences among levels of fixed factors. Tests on the effects of each predictor were performed using *F*-statistics (Crawley 1993). In all cases, we present models based on the data from the end of each experimental period. We analyzed data collected when installing the experimental plots

separately, and in no case was there any significant difference in invertebrate abundance or community parameters among treatments (results not shown). Thus, we are confident that the differences observed among treatments three months later (see results) were not an artefact due to pre-existing patterns in invertebrate distribution.

All invertebrate groups recorded (see Table 8 for details) were considered for analysis, including epibenthic invertebrates. Count data were analyzed using a log link function and negative binomial error distribution (Gray 2005), with the exception of copepods for which a $\log_{10}(x+1)$ transformation and identity link and normal error distribution provided a preferable homoscedastic model. Alternative dependent variables based on the difference recorded in the number of invertebrates from the beginning of an experimental period to the end were rejected because they included negative values to which the negative binomial error distribution can not be applied, and for which no suitable transformations could be found.

Models of rarer taxa did not converge. We thus only present models of counts for the eight most abundant groups: Copepods, the ostracod *Cyprideis torosa*, the shrimp *Palaemonetes varians*, the gastropod *Potamopyrgus antipodarum*, the isopod *Lekanesphaera hookeri*, the amphipod *Corophium orientale*, the corixid *Sigara stagnalis* and chironomid larvae. For amphipods the marginally significant Treatment x Period interaction was kept in the model, because the model without it did not converge. We also present models of mean dry mass for *P. varians*, *P. antipodarum*, *L. hookeri*, *S. stagnalis, C. orientale* and chironomid larvae. Data for copepods and ostracods were not available (see above).

Taxa richness, diversity (Dw), total sample dry weight ($\log_{10}$ transformed) and mean dry mass per sample (square root transformed for *P. antipodarum*, $\log_{10}$ transformed for other taxa) were analyzed using an identity link and normal error distribution. Taxa richness is an integer and has similarities with count data, making a log link function and poisson error distribution an option for this variable. However, alternative models yielded similar results and we chose to present the model with a normal error distribution owing to a preferable distribution of residuals and a greater reduction of deviance (Herrera 2000).

Figures of invertebrate abundance, total dry mass and individual dry masses were plotted using geometric means calculated by $\log_{10}$ transformation of the raw data (or n + 1 for count data) calculation of the arithmetic mean and SE followed by back-transformation, to provide a better representation of skewed data distributions. The only exception was for *P. antipodarum* mass, for which arithmetic means were satisfactory.

RESULTS

Waterbird counts.

Peak numbers of waterbirds in the study site occurred during the post-breeding period, reaching more than 50,000 in November 2001 (Fig. 1). Numbers were also high in winter, and were lowest in March and April (Fig. 1). Numbers of ducks and the largely herbivorous coots followed a similar pattern, peaking in October-November during the post-breeding period. Numbers of flamingos reached a peak in July-August at the height of their breeding season (Fig. 1). The mean density of ducks and coot in Veta la Palma was 2.6 ha^{-1}. The mean density of flamingos was 3.9 ind ha^{-1} (densities were calculated based on the total pond area of 2997 ha). The most abundant waterfowl species recorded (in order of decreasing abundance) were *Fulica atra, Anas platyrhynchos, A. clypeata, A. penelope, A. acuta, A. strepera, Netta rufina* and *Aythya ferina*. The *Anas* dabbling ducks accounted for 96% of the total number of ducks counted. *A. penelope*, *A. clypeata* and *A. acuta* were only present in winter.

Invertebrate community structure

Total sample dry mass was analysed as a measure of the standing crop of invertebrates. Treatment had a highly significant main effect on sample mass (Table 1, Fig. 2), with the highest mass in all-bird exclosures ("no birds"), lowest in controls and intermediate in flamingo exclosures ("no flamingos"). Differences between "no birds" and other treatments were highly significant (Table 2, Fig. 2). There were highly significant differences between periods, and no significant Treatment x Period interaction (Table 1). Sample mass was significantly higher in Jul 01 and Jun 02 than other periods (Table 2, Fig. 2). Water depth had a highly significant partial effect, reflecting an increase in biomass within the water column at greater depths (Table 1). The percentage reduction in total dry mass attributed to "all birds" (i.e. the difference between "all birds" and controls) ranged from 70 to 94% across the four periods. The reduction attributable to flamingos ranged from 2 to 24%, being lowest in Feb02 and highest in Jul01 and Oct01 (Fig. 2).

Both Treatment and Period main effects were highly significant in models of taxonomic richness and of a modified Simpson diversity index, but neither the Treatment x Period interaction nor water depth had significant effects (Table 1). The number of taxa and diversity index were highest in "no birds" and lowest in controls, but only pairwise differences between "no-birds" and other treatments were significant (Table 2, Fig. 2). As with sample mass, richness and diversity were significantly higher in Jul 01 and Jun 02 than other periods (Table 2, Fig. 2).

Invertebrate counts.

Nymphs of the damselfly *Ischnura graellsi*, Coleoptera, Ephydridae larvae and Cladocera were present in our samples (Table 8) but occurred in insufficient numbers to be analysed separately. These four groups together accounted for less than the 1% of the total dry mass and total numbers of invertebrates in samples taken when plots were established at the beginning of the four experimental periods (Table 8). For the eight taxa that were sufficiently numerous to allow individual models (see methods), the Period main effect was highly significant, whilst the Treatment main effect was significant for the isopod *L. hookeri,* the amphipod *C. orientale,* the gastropod *P. antipodarum,* the ostracod *C. torosa* and the corixid *S. stagnalis* (Table 3). Their interaction was significant only for *L. hookeri* (Table 3).

Numbers of *L. hookeri* were highest in "no birds" and lowest in "controls", and all pairwise differences between treatments were significant (Table 4). The order of isopod abundance between periods was Jul01 > Jun02 > Oct01 > Feb02, and all pairwise differences were significant with the exception of Oct01*vs* Feb02 (Table 4, Fig. 3). Post-hoc analysis of the interaction (Table 7) showed that the difference between "no birds" and other treatments was significant in Jun02, whilst *L. hookeri* were significantly more abundant in "no birds" than controls in Oct01.

For the amphipod *C. orientale*, numbers were significantly higher in "no birds" than controls. The order of abundance was Jun02>Jul01>Feb02>Oct01, and all pairwise differences were significant except those between Jul01 and Jun02 or Feb02 (Tables 4, Fig. 3). For *C. torosa*, numbers were significantly lower in "no birds" than in the other treatments, i.e. excluding all birds decreased abundance, contrary to the results for other taxa (Table 4). Between periods, all pairwise differences were highly significant except for Jul01 *vs* Feb02, and the order of ostracod abundance was Jun02 > Feb02 > Jul01 > Oct01 (Table 4, Fig. 3).

For *P. antipodarum* numbers were significantly higher in "no birds" than in controls (Table 4). The order of gastropod abundance between periods for was Jul01 > Jun02 > Feb02 > Oct01, and all pairwise differences were significant with the exception of Oct01 *vs* Feb02 (Table 4, Fig. 3). For the corixid *S. stagnalis*, abundance was significantly higher in 'no-birds' than in other treatments. Between periods all pairwise differences were significant, with an order of abundance of Jun02>Jul01>Oct01>Feb02 (Table 4, Fig. 3).

The order of abundance of copepods between periods was Jun02 > Jul01> Oct01 > Feb02, with all pairwise differences being significant except for Jul01 *vs* Oct01 (Table 4). The abundance of *P.varians* shrimps was significantly higher in Jun02 than in other periods (Table 4, Fig. 3). For abundance of chironomid larvae were significantly higher in Jun02 than in other periods, and was also significantly higher in Oct01 than in Feb02 (Table 4, Fig. 3).

The partial effect of water depth was significant for *C. orientale*, *P. varians*, *S. stagnalis*, chironomids and copepods (Table 3). As depth increased, the abundance of *S. stagnalis* decreased, whilst the abundance of the other four taxa increased (Table 3).

Invertebrate size (mass)

We were able to analyze the mean mass for *P. varians, P. antipodarum, L. hookeri, S. stagnalis*, *C. orientale* and chironomid larvae (see methods). The Treatment main effect was significant for *P. varians,* chironomids and *L. hookeri* (Table 5). The Period main effect was significant for all taxa except for *C. orientale*, whereas the Treatment x Period interaction was only significant for *L. hookeri*. The partial effect of water depth was not significant for any taxon (Table 5).

The mean mass of *P. varians* was significantly higher for "no birds" than for other treatments, and significantly higher in Jul01 and Oct01 than in Feb02 or Jun02 (Table 6, Fig. 4). The mean mass of chironomid larvae was significantly higher in "no flamingos" than in controls (Table 6, Fig. 4). The mean size of chironomid larvae was significantly greater in Feb02 than in Jul01 or Oct01 (Table 6, Fig. 4).

Post-hoc analysis of main effects showed that the mean mass of *L. hookeri* was significantly higher in "no birds" than in other treatments and higher in Jul01 than in Feb02 (Table 6, Fig. 4). Post-hoc analysis of the Treatment x Period interaction showed that significant treatment effects were limited to Jun02 (when mean mass was significantly higher in "no birds" than in controls) and Feb02 (when mean mass was significantly higher in "no birds" than in "no flamingos", Table 7, Fig. 4).

P. antipodarum mean mass was significantly higher in Jun02 than in Oct01 (Table 6, Fig. 4). Mean mass of *S. stagnalis* was significantly higher in Oct01 than in Jul01 or Jun02, and in Feb02 than Jun02 (Table 6, Fig. 4).

DISCUSSION

Most studies on predation by waterbirds have focused on benthic infauna (especially polychaetes and chironomids) and sessile bivalves (Quammen 1984, Glassom and Branch 1997, Hamilton 2000, Marklund & Sandsten 2002). The lack of previous exclosure studies of the effects of waterbirds on invertebrates in the water column may partly be explained by an expectation that the mobility of such invertebrates will make it difficult to detect predation effects. Marklund et al. (2002) studied the effects of waterbirds on a combination of invertebrates taken from sediments and the water column, but found no significant effects. Other studies have shown that waterbird exclusion increases the abundance of epibenthic

invertebrates collected with a benthic corer (Glassom & Branch 1997b, Sutherland et al. 2000, Sherfy & Kirkpatrick 2003). Previous research exploring predation effects by vertebrates on invertebrates in the water column deals mostly with fish, and especially with their effects on plankton communities (see Kerfoot and Sih 1987 for review).

Our results demonstrate that flamingos and especially waterfowl (ducks and coot) can have strong effects on the abundance and community structure of invertebrates in the water column. Elsewhere, we found that both groups had major effects on the benthic infauna and epibenthos collected with a benthic corer during the same experiment (Rodríguez-Pérez & Green unpublished data). Direct predation effects are likely to have made an important contribution to our results. In our study area, analysis of the gut contents of ducks and coots confirmed that they consume chironomid larvae, *S. stagnalis*, *L. hookeri*, *C. orientale*, *C. torosa*, *P. antipodarum* and occasionally *P. varians* (Green & Sánchez 2003, authors unpublished data). Greater flamingos are known to consume ostracods and chironomid larvae (Glassom & Branch 1997 a, Johnson 1997, Rodríguez-Pérez and Green 2006).

However, our results are also likely to reflect the bioturbation (*sensu* Reise 2002) effects of waterbirds and the effects of exclosure on submerged macrophytes, which themselves have a major structuring role on invertebrate communities (see Jeppesen et al. 1998 for review). We found that the frequency of occurrence and biomass of the submerged macrophyte *Ruppia maritima* was greatly increased in our exclosures, especially in the "no birds" treatment (Rodríguez-Pérez and Green 2006). Many of the invertebrate taxa we have recorded are strongly associated with *R. maritima* (Verhoeven 1980), and the abundance of invertebrates has often been shown to be correlated with macrophyte density (Gilinsky 1984, Swisher et al 1998). In most cases, the significant differences we detected between periods indicated that invertebrate abundance was highest in Jul01 and Jun02, as was *R. maritima* biomass (Rodríguez-Pérez and Green 2006). Thus herbivory by ducks and coots, and uprooting of *R. maritima* by feeding flamingos, is likely to have had important indirect effects on the abundance of invertebrates in the water column. For example, in our study area, *L. hookeri* and *P. antipodarum* are largely epiphytes on *Ruppia maritima*. *L. hookeri* is closely associated with macrophytes in brackish lagoons (Healy 1997) and grazes mainly on epiphytic diatoms (Kamermans et al. 2002) but probably also on epibenthic algae and diatoms. However, *R. maritima* is an annual that only provided extensive cover in spring and summer (Rodríguez-Pérez and Green 2006), whereas strong effects of waterbird exclosure on invertebrates were apparent throughout the annual cycle.

Given our experimental design, the strength of treatment effects detected is striking. It might be expected that, given the potential mobility of most of the invertebrate groups included in our study, even strong effects of predation would not be detectable in our exclosures because constant exchange of individuals evens out the density between

exclosures and controls. For example, the failure to detect treatment effects on copepods is not good evidence that waterbirds do not limit copepod density. Both ducks (Euliss et al. 1991, Gaston 1992) and Greater flamingos (Vareschi and Jacobs 1984) are known to prey on copepods, despite their small size. Strong treatment effects for some taxa might partly be a consequence of lower mobility and greater philopatry in some invertebrates than expected from their swimming abilities (Wellborn et al. 1996). It might also reflect a learning ability if some organisms can learn to identify and use areas with reduced predator activity (Boates et al. 1995, Wellborn et al. 1996). Some taxa may well respond to the change in habitat structure, selecting exclosures because they have greater abundance of macrophytes which provide food, or a refuge from other predators such as fish or shrimps. Macrophytes offer a refuge from predation (Diehl and Kornijow 1998) and predator success tends to decline as macrophyte density increases (Gilinsky 1984, Swisher et al 1998, but see Warfe and Barmuta 2004). For all these reasons, it is difficult to interpret our treatment effects and associate them to direct effects of predation.

Our results on the strong effects of waterbirds are particularly striking as fish were abundant in our study area and were not affected by the exclosures. Fish are traditionally considered to be far more relevant to invertebrate communities than waterbirds (E.g. Wetzel 2001). Compensatory predation by fish of invertebrates in exclosures may even have reduced our ability to detect bird effects (Marklund et al. 2002).

In all community traits examined we found significant treatment effects. Waterfowl had significant additive negative effects on the diversity, taxa richness and total dry mass of the invertebrates in the water column, although it seems likely that flamingos made an important contribution to the total exclusion effect (Fig 2). The lack of significant treatment x period interactions in our models of community parameters suggests that the fluctuations in abundance of waterbirds had relatively little influence on the strength of their effects. However, the percentage reduction, with untransformed data, in total dry mass was greatest in Oct01 and Feb02 (ca 90%), when the abundance of waterfowl peaked, and when the difference in abundance between waterfowl and flamingos was greatest (Fig. 1). *P. varians* was the major contributor to total dry mass, its mass being one order of magnitude higher than that of any other taxon (see Table 8). Thus the changes in total mass are closely related to changes in the biomass of *P. varians*.

In shallow lakes (*sensu* Scheffer 1998) such as our study area, distinctions between functional groups of invertebrates such as benthos, nekton, plankton and epiphytes are somewhat unsatisfactory and artificial. The strength of interactions between benthic and pelagic communities is strongly affected by lake size (Vadeboncoeur et al. 2002) and the distinction between these communities becomes blurred in shallow systems. Taxa such as *C. orientale*, *C. torosa* and chironomid larvae can burrow into sediments, but also spend time

swimming freely in the water column (personal observation). However, some authors have supported the differentiation between benthic and non-benthic macroinvertebrates in shallow systems such as ours (van de Meutter et al. 2005, Diehl and Kornijow 1998), based on different food sources, predators and seasonal dynamics. There is important overlap between the benthic fauna studied in our experiments using a benthic corer (Rodríguez-Pérez and Green unpublished data), and the invertebrates sampled in the current study. *P. antipodarum*, *C. torosa* and chironomid larvae were abundant in both sets of samples. However, our benthic samples core were dominated by polychaetes, whereas *P. varians*, *S. stagnalis* and copepods were almost absent.

The abundance of *P. varians*, *C. orientale*, chironomid larvae and copepods in our samples from the water column increased with water depth. This may largely be explained by the increase in sample volume at a greater depth, and by a positive correlation between *R. maritima* biomass and depth (authors unpublished). In contrast, the abundance of *S. stagnalis* decreased with depth. Similar correlations with depth for corixids, ostracods and copepods have been recorded in other brackish lakes in Mediterranean Spain (Fuentes, 2005).

The mean mass of *P. varians* and *L. hookeri* was higher in all-bird exclosures than in other treatments. This may reflect indirect effects related to the abundance of submerged macrophytes. However, it is also possible that ducks feed selectively on larger *L. hookeri*, as has been observed previously for amphipods (Batzer et al. 1993). The numbers of isopods were also higher in all-bird exclosures than other treatments, and were also increased by excluding flamingos. This could be due to predation by flamingos, but might also be due to a greater food supply for *L. hookeri* in flamingo exclosures than in controls, since disturbance of sediments by flamingos is likely to inhibit the growth of algae and diatoms on the sediment surface and on organic litter. Similarly, the increase in mean mass of chironomid larvae recorded when excluding flamingo may be a result of either predation or bioturbation.

It is unlikely that the differences in size of *P. varians* between treatments are directly caused by predation, since this shrimp is a fast swimmer well equipped to escape waterbirds, and because the abundance of shrimps was not significantly increased in exclosures. Indeed, shrimp numbers seemed to be lower in all-bird exclosures in Jun02 (Fig 2). Such a reduction in density but increase in size in exclosures is usually due to adult-larval competition (Woodin 1976, Olafsson et al. 1994), and it is possible that larger shrimps prefer the undisturbed, well-vegetated microhabitats offered by all-bird exclosures, and displace smaller shrimps from them. *Palaemonetes* species are omnivorous (Kneib 1988, Irvine et al. 1993, Guerao and Ribera 1996, and seem likely to prey on some of the other taxa included in our study.

An increase in predation risk from larger shrimps and more corixids in all-bird exclosures may explain why *C. torosa* exhibited an indirect effect of waterbird predation (Sih et al. 1985, Wootton 2002) by decreasing their density in all-bird exclosures compared to other treatments. Such indirect effects are usually explained by compensatory predation or by competition (Kneib 1988, Wilson 1991, Thrush 1999, Hamilton 2000). *S. stagnalis* and *P. varians* are both likely to prey on ostracods (Kneib 1988, Guerao and Ribera 1996, Irvine et al. 1993, Barahona et al. 2005), whereas *C. torosa* may also compete with and be displaced by the amphipod *C. orientale* (Modig et al. 2000, Ejdung and Elmgren 1998).

In conclusion, we have found strong evidence for a major role of waterbirds in structuring the invertebrate community in the water column of shallow lakes. The densities of waterbirds present in our study were relatively low compared with other studies of their effects on aquatic ecosystems (Marklund 2002), suggesting that this major role for waterbirds is not a special feature of our study area. Both waterfowl and greater flamingos had significant effects, although the effects of flamingos were much weaker than those recorded for benthic infauna (Rodríguez-Pérez and Green unpublished). The relative roles of predation, herbivory and bioturbation in causing these effects remains to be explored in future studies. Further studies should also aim to establish to what extent this structuring role for birds is consistent between systems.

Acknowledgements:

Esther Macarro provided invaluable assistance during the field and processing work. We also thank Mª del Mar Delgado for her help at the lab, and Jordi Figuerola, Carlos M. Herrera, Pedro Jordano and Antonio Manzaneda for statistical advice. Ángel Baltanás, José Manuel Guerra, Andrés Millán, and Juan Carlos Pérez assisted with taxonomic identifications. Pesquerías Isla Mayor S.A. and the Department of Environment (Consejería de Medio Ambiente) of the Andalusian Autonomous Government gave us permission to work in Veta la Palma. This work was funded by the Consejería de Medio Ambiente. Héctor Rodríguez Pérez had a PhD grant from the CSIC-I3P programme funded by the European Social Fund. Aerial count data were provided by the Equipo de Seguimiento de Procesos of Doñana Biological Station.

References:

Argano, R. 1979. Guide per il riconoscimiento delle specie animali delle acque interne italiane 5. Isopodi. Consiglio Nazionale delle Ricerche.

Askew, R.R. 1988. The dragonflies of Europe. 291 pp. Harley Books.

Barahona, J., A. Millan, and J. Velasco. 2005. Population dynamics, growth and production of *Sigara selecta* (Fieber, 1848) (Hemiptera, Corixidae) in a Mediterranean hypersaline stream. Freshwater Biology 50:2101-2113.

Batzer, D.P., M. McGee, V.H. Resh, and R.R. Smith. 1993. Characteristics of invertebrates consumed by mallards and prey response to wetland flooding schedules. Wetlands 13(1):41-49.

Boates, J. S., M. Forbes, M. Zinck, and N. McNeil. 1995. Male amphipods (*Corophium volutator*) show flexible behaviour in relation to risk of predation by sandpipers. Ecoscience 2:123-128.

Botto, F., O. O. Iribarne, M. M. Martinez, K. Delhey, and M. Carrete. 1998. The effect of migratory shorebirds on the benthic species of three southwestern Atlantic Argentinean estuaries. Estuaries 21:700-709.

Castello, J. 1986. Contribución al conocimiento biológico de los crustáceos del litoral Catalano-Balear. PhD Thesis. Barcelona University.

Cooper, S. D., D. W. Winkler, and P. H. Lenz. 1984. The Effect of Grebe Predation on a Brine Shrimp Population. Journal of Animal Ecology 53:51-64.

Crawley, MJ. 1993. GLIM for ecologists. Blackwell Science.

Crowder, L.B., E.W. McCollum, and T.H. Martin. 1998. Changing perspectives on food web interactions in lake littoral zones. Pages 240-249 in Jeppensen, E., M. Sondergaard, M. Sondergaard, and K. E. Christoffersen. 1998. The structuring role of submerged macrophytes in lakes. Spring-Verlag, New York.

deSzalay, F. A., and V. H. Resh. 1997. Responses of wetland invertebrates and plants important in waterfowl diets to burning and mowing of emergent vegetation. Wetlands 17:149-156.

Diehl, S. 1995. Direct and Indirect Effects of Omnivory in a Littoral Lake Community. Ecology 76:1727-1740.

Diehl, S., and R. Kornijów. 1998. Influence of submerged macrophytes on trophic interactions among fish and macroinvertebrates. Pages24-46 in Jeppensen, E., M. Sondergaard, M. Sondergaard, and K. E. Christoffersen. 1998. The structuring role of submerged macrophytes in lakes. Spring-Verlag, New York.

Ejdung G., and R. Elmgren. 1998. Predation on newly settled bivalves by deposit-feeding amphipods: a Baltic Sea case study. Marine Ecology Progress Series 168:87-94.

Euliss, N. H., R. L. Jarvis, and D. S. Gilmer. 1991. Feeding Ecology of Waterfowl Wintering on Evaporation Ponds in California. Condor 93:582-590.

Frisch, D., H. Rodríguez-Pérez, and A.J. Green. In press. Invasion of artificial ponds in Doñana Natural Park, southwest Spain, by an exotic estuarine copepod. Aquatic conservation: Marine and Freshwater ecosystem. DOI: 10.1002/aqc.718.

Fuentes, C. 2005. Ecología de la Cerceta Pardilla (*Marmaronetta angustirostris*) y de la Malvasía cabeciblanca (Oxyura leucocephala) en los humedales del Baix Vinalopó, Alicante. Ph D Thesis. Alicante University.

Gaston, G.R. 1992. Green-Winged Teal Ingest Epibenthic Meiofauna. Estuaries 15:227-229.

Ghetti, P.F., and K. Mc Kenzie. 1981. Guide per il riconoscimiento delle specie animali delle acque interne italiane 11. Ostracodi. Consiglio Nazionale delle Ricerche.

Gilinsky, E. 1984. The Role of Fish Predation and Spatial Heterogeneity in Determining Benthic Community Structure. Ecology 65:455-468.

Glassom, D., and G. M. Branch. 1997 a. Impact of predation by greater flamingos *Phoenicopterus ruber* on the macrofauna of two southern African lagoons. Marine Ecology Progress Series 149:1-12.

Glassom, D., and G. M. Branch. 1997 b. Impact of predation by greater flamingos Phoenicopterus ruber on the meiofauna, microflora, and sediment properties of two southern African lagoons. Marine Ecology Progress Series 150:1-10.

Gray, B.R. 2005. Selecting a distributional assumption for modelling relative densities of benthic macroinvertebrates. Ecological Modelling 185(1):1-12.

Green, A.J., and M.I. Sánchez. 2003. Spatial and temporal variation in the diet of Marbled Teal Marmaronetta angustirostris in the western Mediterranean. Bird Study 50:153-160.

Guerao, G., and C. Ribera. 1996. Locomotor activity patterns and feeding habitats in the prawn Palaemon serratus (Pennant, 1777) (Decapoda, Palaemonidae)in the Alfacs bay, Ebro delta, Spain. Crustaceana:101-112.

Hamilton, D. J. 2000. Direct and indirect effects of predation by Common Eiders and abiotic disturbance in an intertidal community. Ecological Monographs 70:21-43.

Hamilton, D.J., and T.D. Nudds. 2003. Effects of predation by common eiders (*Somateria mollissima*) in an intertidal rockweed bed relative to an adjacent mussel bed. Marine Biology 142(1): 1-12.

Healy, B. 1997. long-term changes in a brackish lagoon, Lady's Island lake, South-East Ireland. Biology and Environment: Proceedings of the Royal Irish Academy 97 B:33-51.

Herrera, C.M. 2000. Flower-to-seedling consequences of different pollination regimes in an insect-pollinated shrub. Ecology 81(1):15-29.

Irvine, K., B. Moss, M. Bales, and D. Snook. 1993. The Changing Ecosystem of a Shallow, Brackish Lake, Hickling Broad, Norfolk, Uk .1. Trophic Relationships with Special Reference to the Role of Neomysis-Integer. Freshwater Biology 29:119-139.

Jansson, A. 1986. The corixidae (Heteroptera) of Europe and some adjacent regions. 94 pp. Acta Entomologica Fennica 47.

Jeppensen, E., M. Sondergaard, M. Sondergaard, and K. E. Christoffersen. 1998. The structuring role of submerged macrophytes in lakes. Spring-Verlag, New York.

Johnson, A. R. 1997. Long-term studies and conservation of Greater Flamingos in the Camargue and Mediterranean. Colonial Waterbirds 20:306-315.

Kamermans, P., E.-J. Malta, J. M. Verschuure, L. Schrijvers, L. F. Lentz, and A. T. Lien. 2002. Effect of grazing by isopods and amphipods on growth of Ulva spp. (Chlorophyta). Aquatic Ecology 36:425-433.

Kear, J. (Ed.). 2005. Ducks, geese and swans. Oxford University Press, Oxford.

Kerfoot, W.C., and A. Sih. 1987. Predation. Direct and indirect impacts on aquatic communities. 386 pp. University Press of New England.

Kneib, R. T. 1988. Testing for Indirect Effects of Predation in an Intertidal Soft-Bottom Community. Ecology 69:1795-1805.

Lodge, D.M., G. Cronin, E. van Donk, and A.J. Froelich. 1998. Impact of hervibory on plant standing crop: comparisons among biomes, between vascular and nonvascular plants, and among freshwater herbivore taxa. Pages 149-174 in Jeppensen, E., M. Sondergaard, M. Sondergaard, and K. E. Christoffersen. 1998. The structuring role of submerged macrophytes in lakes. Spring-Verlag, New York.

Magurran, A.E. 1988. Ecology diversity and its measurements. 179pp. Croom Helm. London

Marklund, O., H. Sandsten, L.-A. Hansson, and I. Blindow. 2002. Effects of waterfowl and fish on submerged vegetation and macroinvertebrates. Freshwater Biology 47:2049-2059.

Marklund, O., and H. Sandsten. 2002. Reduction of benthic macroinvertebrates due to waterfowl foraging on submerged vegetation during autumn migration. Aquatic Ecology 36:541-547.

Mc Cullagh, P. and Nelder, J.A. 1989. Generalized linear models. Chapman and Hall.

Modig, H., W. J. van de Bund, and E. Olafsson. 2000. Uptake of phytodetritus by three ostracod species from the Baltic Sea: effects of amphipod disturbance and ostracod density. Marine Ecology-Progress Series 202:125-134.

Olafsson, E. B., C. H. Peterson, and W. G. Ambrose. 1994. Does Recruitment Limitation Structure Populations and Communities of Macroinvertebrates in Marine Soft

Sediments - the Relative Significance of Presettlement and Postsettlement Processes. Oceanography and Marine Biology, Vol 32 32:65-109.

Ortiz, M., and A. Jimeno. 2001. Guía ilustrada para la identificación de las familias y los géneros de los anfípodos del suborden gammaridea de la península Ibérica. Graellsia 57(2):3-93.

Pöysä, H. 1983. Morphology-mediated niche organization in a guild of dabbling ducks. Ornis Scandinavica 14:317-326.

Quammen, M. L. 1984. Predation by Shorebirds, Fish, and Crabs on Invertebrates in Intertidal Mudflats - an Experimental Test. Ecology 65:529-537.

Reise, K. 2002. Sediment mediated species interactions in coastal waters. Journal of Sea Research, 48, 127-141.

Richardson, W. B., S. A. Wickham, and S. T. Threlkeld. 1990. Foodweb Response to the Experimental Manipulation of a Benthivore (Cyprinus-Carpio), Zooplanktivore (Menidia-Beryllina) and Benthic Insects. Archiv Fur Hydrobiologie 119:143-165.

Rodríguez-Pérez, H., and A.J. Green. 2006. Waterbird impacts on widgeongrass Ruppia maritime in a Mediterranean wetland: comparing bird groups and seasonal effects. Oikos 112: 525-534.

Rubega, M., and C. Inouye. 1994. Prey Switching in Red-Necked Phalaropes Phalaropus-Lobatus - Feeding Limitations, the Functional-Response and Water Management at Mono-Lake, California, USA. Biological Conservation 70:205-210.

Ruffo, S. 1982. The Amphipoda of the Mediterranean. Mémoires de l'Institut Océanographique 13. Fondation Albert I Prince de Monaco.

SAS Institute Inc. 2005. SAS GLIMMIX.

Scheffer, M. 1998. Ecology of shallow lakes. 357 pp. Kluwer Academic Publisher, Dordrecht.

Sherfy, M. H., and R. L. Kirkpatrick. 2003. Invertebrate response to snow goose herbivory on moist-soil vegetation. Wetlands 23:236-249.

Shuterland, T.F., P.C.F. Sheperd, and R.W. Elner. 2000. Predation on meiofaunal and macrofaunal invertebrates by western sandpipers (*Calidris mauri*): evidences of dual foraging mode. Marine Biology 137: 983-993.

Sih, A., P. Crowley, M. Mcpeek, J. Petranka, and K. Strohmeier. 1985. Predation, Competition, and Prey Communities - a Review of Field Experiments. Annual Review of Ecology and Systematics 16:269-311.

Swisher, B.J., D.A. Soluk, and D.H. Wahl. 1998. Non-additive predation in littoral habitats: influences of habitat complexity. Oikos 81(1):30-37.

Tachet, H., P. Richoux, M. Bournaud and P. Usseglio-Polaterra. 2003. Invertébrés d'eau douce- Systématique, biologie, ecologie. 587 pp. CNRS Editions, Paris.

Thrush, S. E. 1999. Complex role of predators in structuring soft-sediment macrobenthic communities: Implications of changes in spatial scale for experimental studies. Australian Journal of Ecology 24:344-354.

Vadeboncoeur, Y., M. J. Vander Zanden, and D. M. Lodge. 2002. Putting the lake back together: Reintegrating benthic pathways into lake food web models. Bioscience 52:44-54.

van de Meutter, F., R. Stoks, and L. De Meester. 2005. The effect of turbidity state and microhabitat on macroinvertebrate assemblages: a pilot study of six shallow lakes. Hydrobiologia 542:379-390.

Vareschi, E., and J. Jacobs. 1984. The Ecology of Lake Nakuru (Kenya) .5. Production and Consumption of Consumer Organisms. Oecologia 61:83-98.

Verhoeven, J. T. A. 1980. The Ecology of Ruppia-Dominated Communities in Western-Europe .2. Synecological Classification - Structure and Dynamics of the Macroflora and Macrofauna Communities. Aquatic Botany 8:1-85.

Warfe, D. M., and L. A. Barmuta. 2004. Habitat structural complexity mediates the foraging success of multiple predator species. Oecologia 141:171-178.

Wellborn, G. A., D. K. Skelly, and E. E. Werner. 1996. Mechanisms creating community structure across a freshwater habitat gradient. Annual Review of Ecology and Systematics 27:337-363.

Wilson, W. H. 1991. Competition and Predation in Marine Soft-Sediment Communities. Annual Review of Ecology and Systematics 21:221-241.

Woodin, S. A. 1976. Adult-Larval Interactions in Dense Infaunal Assemblages - Patterns of Abundance. Journal of Marine Research 34:25-41.

Wootton, J. T. 2002. Indirect effects in complex ecosystems: recent progress and future challenges. Journal of Sea Research 48:157-172.

	Total weight						Richness						Diversity (Dw)					
FACTOR	**Estimate**	**SE**	**df_N**	**df_D**	**F**	**p**	**Estimate**	**SE**	**df_N**	**df_D**	**F**	**p**	**Estimate**	**SE**	**df_N**	**df_D**	**F**	**p**
Intercept	-0.87	0.14					7.38	0.54					1.94	0.19				
Treatment (T)			2	164	40.31	<0.0001			2	164	8.50	0.0003			2	164	9.03	0.0002
C	-0.62	0.07					-0.78	0.19					-0.35	0.08				
F	-0.53	0.07					-0.44	0.19					-0.2	0.08				
Date (D)			3	164	23.35	<0.0001			3	164	46.64	<0.0001			3	164	24.71	<0.0001
Jul 01	-0.09	0.1					0.44	0.26					-0.14	0.11				
Oct 01	-0.5	0.09					-1.96	0.25					-0.7	0.11				
Feb 02	-0.6	0.09					-1.43	0.23					-0.7	0.1				
Depth	0.02	0.005	1	164	19.03	<0.0001	0.01	0.02	1	164	0.49	0.5	-0.002	0.007	1	164	0.09	0.8
TxD						n.s.						n.s.						n.s.

Table 1.- Summary of generalized linear models testing the main factors treatment (three levels), date (four levels) and depth and the Treatment x Date interaction on community traits: taxonomic richness, diversity (weighted Simpson Index Dw), and total dry mass (g). When the interaction was not significant ($p>0.05$) it was removed from the analysis. Pond and plot were included as random factors using the GLIMMIX procedure with an identity link function and normal error distribution for richness, diversity and total weight (log_{10} transformed). See methods for details. The all-bird exclosure treatment and date Jun 02 were aliased. C=controls, F= flamingo exclosures.

	FACTOR	Total weight		Richness		Dw	
		T_{164}	p	T_{164}	p	T_{164}	p
Treatment	C vs F	-1.17	0.5	-1.83	0.16	-1.78	0.2
	C vs A	-8.28	<0.0001	-4.11	0.002	-4.32	0.0001
	F vs A	-7.21	<0.0001	-2.36	0.05	-2.53	0.03
Date	Jul 01 vs Oct 01	4.94	<0.0001	10.24	<0.0001	5.61	<0.0001
	Jul 01 vs Feb 02	5.82	<0.0001	8.01	<0.0001	5.20	<0.0001
	Jul 01 vs Jun 02	-0.93	0.8	1.71	0.3	-1.27	0.6
	Oct 01 vs Feb 02	0.96	0.8	-2.43	0.08	-0.43	0.9
	Oct 01 vs Jun 02	-5.6	<0.0001	-7.93	<0.0001	-6.63	<0.0001
	Feb 02 vs Jun 02	-6.75	<0.0001	-6.14	<0.0001	-6.58	<0.0001

Table 2.- Post-hoc least-squared means tests comparing pairwise differences among levels of the main effects treatment (three levels) and date (four levels) that were significant when analysing taxonomic richness, diversity (weighted Simpson Index Dw) and total dry mass (see Table 1). P-values were adjusted with Tukey tests. C= controls, F=flamingo exclosures, A= all-bird exclosures.

	Lekanesphera hookeri						*Corophium orientale*						*Palaemonetes varians*						*Potamopyrgus antipodarum*					
FACTOR	**Estimate**	**SE**	**df_N**	**df_D**	***F***	**p**	**Estimate**	**SE**	**df_N**	**df_D**	***F***	**p**	**Estimate**	**SE**	**df_N**	**df_D**	**F**	**p**	**Estimate**	**SE**	**df_N**	**df_D**	**F**	**p**
Intercept	3.9	0.6					-0.8	1.16					2.56	0.52					1.56	0.73				
Treatment (T)			2, 164		28.51	<0.0001			2, 158		3.51	0.03			2, 164		1.64	0.2			2, 164		4.93	0.008
C	-2.59	0.45					-1.19	0.5					-0.42	0.24					-0.53	0.17				
F	-1.94	0.43					-0.87	0.5					-0.13	0.23					-0.35	0.17				
Date (D)			3, 164		104	<0.0001			1, 158		30.81	<0.0001			3, 164		13.27	<0.0001			3, 164		26.68	<0.0001
Jul 01	1.87	0.45					-1.51	0.58					-1.59	0.31					0.86	0.22				
Oct 01	-1.56	0.45					-3.25	0.59					-1.57	0.3					-0.79	0.22				
Feb 02	-2.39	0.46					-2.02	0.57					-1.63	0.3					-0.62	0.21				
Depth	-0.02	0.02	1, 164		1.05	0.3	0.13	0.03	1, 158		16.49	<0.0001	0.08	0.02	1, 164		15.63	0.0001	0.03	0.02	1, 164		3.79	0.053
TxD			6, 164		2.21	0.04			2, 158		2.12	0.054						n.s.						n.s.
C x Jul 01	1.86	0.6					1.24	0.75																
C x Oct 01	0.15	0.7					-1.16	0.9																
C x Feb 02	1.03	0.7					-1.45	0.71																
F x Jul 01	1.28	0.59					-1.43	0.75																
F x Oct 01	0.68	0.64					-0.3	0.82																
F x Feb 02	1.18	0.66					0.81	0.72																
	Copepods						***Cyprideis torosa***						***Sigara stagnalis***						**Chironomids**					
FACTOR	**Estimate**	**SE**	**df_N**	**df_D**	***F***	**p**	**Estimate**	**SE**	**df_N**	**df_D**	***F***	**p**	**Estimate**	**SE**	**df_N**	**df_D**	**F**	**p**	**Estimate**	***SE***	**df_N**	**df_D**	**F**	**P**
Intercept	0.65	0.33					6.7	0.39					8.01	0.8					0.99	0.21				
Treatment (T)			2, 164		0.00	0.9			2, 164		14.92	<0.0001			2, 164		5.28	0.006			2, 164		0.44	0.6
C	-0.006	0.1					0.71	0.14					-0.64	0.21					0.003	0.09				
F	-0.004	0.1					0.62	0.14					-0.5	0.2					-0.07	0.09				
Date (D)			3, 164		26.66	<0.0001			3, 164		63.25	<0.0001			1, 164		116.84	<0.0001			3, 164		25.45	<0.0001
Jul 01	-0.36	0.14					-1.67	0.19					-1.72	0.27					-0.83	0.12				
Oct 01	-0.55	0.13					-2.52	0.18					-3.33	0.27					-0.63	0.12				
Feb 02	-1.1	0.12					-1.38	0.17					-4.12	0.26					-0.92	0.11				
Depth	0.07	0.01	1, 164		51.18	<0.0001	-0.004	0.01	1, 164		0.08	0.8	-0.13	0.02	1, 164		32.09	<0.0001	0.02	0.008	1, 164		4.14	0.04
TxD						n.s.						n.s.						n.s.						n.s.

Table 3.- Summary of generalized linear models testing the main effects treatment (three levels), date (four levels) and depth and the Treatment x Date interaction on invertebrate counts. Pond and plot were included as random factors using the GLIMMIX procedure with a log link function and negative binomial distribution, except for copepods for which a $\log_{10}$ transformation and identity link function and normal error distribution were used. See methods for details. The all-bird exclosure treatment and date Jun 02, and all combinations with at least one of these levels were aliased. C= controls, F= flamingo exclosures.

		Copepods		*C. torosa*		*P. varians*		*P. antipodarum*		*L. hookeri*		*C. orientale*		*S. stagnalis*		Chironomids	
	FACTOR	T_{164}	**p**	T_{164}	**p**	T_{164}	**p**	T_{164}	**p**	T_{164}	**p**	T_{158}	**p**	T_{164}	**p**	T_{164}	**p**
Treatment	C vs F			0.64	0.8			-1.02	0.6	-2.61	0.03	-1.39	0.3	-0.71	0.8		
	C vs A			5.01	<0.0001			-3.07	0.007	-7.26	<0.0001	-2.64	0.02	-3.07	0.007		
	F vs A			4.46	<0.0001			-2.12	0.09	-5.07	<0.0001	-1.37	0.4	-2.45	0.04		
Date	Jul 01 vs Oct 01	1.56	0.4	4.85	<0.0001	-0.08	0.9	8.10	<0.0001	14.36	<0.0001	7.92	<0.0001	10.11	<0.0001	-1.83	0.3
	Jul 01 vs Feb 02	5.68	<0.0001	-1.68	0.3	0.13	0.9	7.31	<0.0001	15.43	<0.0001	1.95	0.2	12.68	<0.0001	0.76	0.9
	Jul 01 vs Jun 02	-2.60	0.05	-8.73	<0.0001	-5.07	<0.0001	3.88	<0.0001	9.61	<0.0001	-1.67	0.3	-2.71	0.04	-6.86	<0.0001
	Oct 01 vs Feb 02	4.46	<0.0001	-7.06	<0.0001	0.23	0.9	-0.88	0.8	1.19	0.6	-6.60	<0.0001	2.91	0.02	2.76	0.03
	Oct 01 vs Jun 02	-4.20	0.0003	-13.71	<0.0001	-5.18	<0.0001	-3.59	0.002	-3.99	<0.0001	-9.02	<0.0001	-12.49	<0.0001	-5.42	<0.0001
	Feb 02 vs Jun 02	-8.57	<0.0001	-7.97	<0.0001	-5.63	<0.0001	-3.01	0.02	-5.43	<0.0001	-3.46	0.004	-15.66	<0.0001	-8.28	<0.0001

Table 4.- Post-hoc least-squared means test comparing pairwise differences among levels of the main effects treatment (three levels) and date (four levels) that were significant in analyses of invertebrate counts (see Table 3). Post-hoc tests were not performed when main effects were not significant. P-values were adjusted with Tukey tests. C= controls, F= flamingo exclosures, A= all-bird exclosures.

FACTOR	*Palaemonetes varians* (w)						*Potamopyrgus antipodarum* (w)						Chironomids(w)						*Lekanesphera hookeri* (w)					
	Estimate	**SE**	**df_N**	**df_D**	**F**	**p**	**Estimate**	**SE**	**df_N**	**df_D**	**F**	**p**	**Estimate**	**SE**	**df_N**	**df_D**	**F**	**p**	**Estimate**	**SE**	**df_N**	**df_D**	**F**	**p**
Intercept	-2.94	0.28					-0.65	0.06					-4.07	0.14					-3.32	0.16				
Treatment (T)			2, 148		24.84	<0.0001			2, 140		0.05	0.9			2, 109		3.13	0.05			2, 95		6.34	0.003
C	-0.99	0.15					0.008	0.04					-0.16	0.1					-0.74	0.19				
F	-0.79	0.15					-0.003	0.04					0.07	0.1					-0.1	0.18				
Date (D)			3, 148		5.83	0.0009			3, 140		23.63	0.04			3, 109		5.15	0.002			3, 95		3.86	0.01
Jul 01	0.48	0.2					-0.07	0.05					-0.21	0.12					-0.19	0.17				
Oct 01	0.51	0.18					-0.13	0.05					-0.20	0.11					-0.42	0.18				
Feb 02	-0.08	0.18					-0.04	0.04					0.18	0.11					-0.17	0.19				
Depth	0.02	0.01	1, 148		3.46	0.07	0.002	0.002	1, 140		0.86	0.4	-0.003	0.006	1, 109		0.3	0.6	-0.001	0.007	1, 95		0.04	0.9
TxD						n.s.						n.s.						n.s.			6, 95		3.46	0.004
C x Jul 01																			0.73	0.25				
C x Oct 01																			0.52	0.33				
C x Feb 02																			0.24	0.31				
F x Jul 01																			-0.04	0.24				
F x Oct 01																			0.18	0.28				
F x Feb 02																			-0.68	0.27				

FACTOR	*Sigara stagnalis* (w)						*Corophium orientale* (w)					
	Estimate	**SE**	**df_N**	**df_D**	**F**	**P**	**Estimate**	**SE**	**df_N**	**df_D**	**F**	**p**
Intercept	-3.83	0.19					-4.2	0.26				
Treatment (T)			2, 114		0.44	0.6			2, 76		0.81	0.4
C	-0.03	0.07					-0.08	0.1				
F	-0.04	0.07					-0.12	0.09				
Date (D)			3, 114		6.56	0.0004			3, 76		0.67	0.6
Jul 01	0.08	0.09					0.03	0.13				
Oct 01	0.34	0.09					-0.07	0.15				
Feb 02	0.33	0.1					0.13	0.12				
Depth	0.01	0.007	1, 114		1.99	0.16	-0.006	0.1	1, 76		0.31	0.4
TxD						n.s.						n.s.

Table 5.- Summary of generalized linear models testing the main effects treatment (three levels) and date (four levels), and depth and treatment x date interaction on invertebrate individual mean dry mass (g, $\log_{10}$ transformed except for *P. antipodarum* which was square root transformed). When the interaction was not significant ($p>0.05$) it was removed from the analysis. Pond and plot were included as random factors using GLIMMIX procedure with an identity link and normal error distribution. See methods for details. All-birds exclosures treatment and date Jun 02, and all combinations with at least one of these levels, were aliased. C= controls, F= flamingo exclosures.

		P. varians (w)		*P. antipodarum* (w)		Chironomids (w)		*L. hookeri* (w)		*S. stagnalis* (w)	
	FACTOR	**T_{148}**	**p**	**T_{140}**	**p**	**T_{109}**	**p**	**T_{95}**	**p**	**T_{114}**	**p**
Treatment	C vs F	-1.39	0.4			-2.42	0.04	-1.17	0.5		
	C vs A	-6.62	<0.0001			-1.69	0.2	-3.35	0.003		
	F vs A	-5.42	<0.0001			0.67	0.8	-2.40	0.05		
Date	Jul 01 vs Oct 01	-0.21	0.9	1.44	0.47	-0.09	0.99	1.75	0.3	-2.85	0.03
	Jul 01 vs Feb 02	3.02	0.02	-0.66	0.9	-3.37	0.006	3.07	0.02	-2.45	0.07
	Jul 01 vs Jun 02	2.43	0.08	-1.55	0.4	-1.81	0.3	0.34	0.99	0.84	0.8
	Oct 01 vs Feb 02	3.51	0.003	-2.07	0.2	-3.35	0.006	0.97	0.77	0.14	0.9
	Oct 01 vs Jun 02	2.80	0.03	-2.80	0.03	-1.76	0.3	-1.31	0.56	3.66	0.002
	Feb 02 vs Jun 02	-0.45	0.9	-0.97	0.8	1.58	0.4	-2.54	0.06	3.31	0.007

Table 6.- Post-hoc least squared means test comparing pairwise differences among levels of the main effects treatment (three levels) and date (four levels) when these were significant in analyses of invertebrate individual mean mass (see Table 5). Post-hoc tests were not performed when main effects were not significant. P-values were adjusted with Tukey tests.C= controls, F= flamingo exclosures, A= all-bird exclosures.

		Jul 01		Oct 01		Feb 02		Jun 02	
		t	p	T	p	t	p	T	p
L. hookeri df=164	C vs. F	-0.13	1.0	-2.05	0.7	-1.39	0.9	-1.41	0.9
	C vs. A	-1.74	0.9	-4.44	<.0001	-2.84	0.2	-5.74	<.0001
	F vs. A	-1.62	0.9	-2.70	0.2	-1.54	0.9	-4.48	0.0009
***L. hookeri* (w)** df=95	C vs. F	0.76	0.5	-1.04	0.9	1.11	0.9	-3.13	0.09
	C vs. A	-0.06	1.0	-0.82	0.9	-2.10	0.6	-3.85	0.01
	F vs. A	-0.84	0.9	0.38	1.0	-3.79	0.01	-0.53	1.0

Table 7.- Post-hoc least-squared means test on significant interactions among treatment and date, comparing pairwise differences among treatments (factor of three levels) for different dates (factor of four levels) for *L. hookeri* counts and mean dry mass. P-values were adjusted with Tukey tests. (w)= body size model, C= controls, F= flamingo exclosures, A= all-bird exclosures

Class	Groups identified	Apr 01		Jul 01		Jul 01		Oct01		Nov01		Feb 02		Mar 02		Jun 02	
		N	B (g/m^2)	N	B (g/m^2)	N	B (g/m^2)	N	B (g/m^2)	N	B (g/m^2)	N	B (g/m^2)	N	B (g/m^2)	N	B (g/m^2)
Maxillopoda	O. Cyclopoida F. Cyclopoida *Halicyclops magniceps* (Lilljeborg 1853) *Halicyclops neglectus* (Kiefer 1935) *Halicyclops rotundipes* (Kiefer 1935) O. Calanoida F. Acartiidae Acartia tonsa (Dana 1948) F. Pseudodiaptomidae *Calanipeda aquae-dulcis* (Kritschagin 1873) O. Harpacticoida F. Cletodidae *Cletocamptus* cf. *Retrogressus* (Shmankevich 1875)	11697	*1.924*	32155	*5.287*	44894	*7.389*	38143	*6.274*	3855	*0.634*	3231	*0.532*	4772	*0.783*	21706	*3.567*
Branchiopoda	O. Anomopoda F. Moinidae *Moina brachiata* (Jurine 1820) F. Daphniidae *Daphnia* (*Ctenodaphnia*) magna (Straus 1820)	0	*0*	59	*~0*	518	*0.002*	0	*0*	0	*0*	0	*0*	1	*~0*	25	*~0*
Ostracoda	O. Podocopa F. Cytherideidae *Cyprideis torosa* (Jones 1857)	20524	*39.490*	21933	*42.038*	10533	*20.191*	5290	*10.127*	9239	*17.707*	11925	*22.866*	13752	*26.369*	41248	*78.981*
Gastropoda	O. Neotaeniglossa F. Hydrobiidae *Potamopyrgus antipodarum* (Gray 1843)	582	*4.584*	2782	*36.051*	3698	*59.847*	711	*16.831*	461	*7.089*	526	*16.240*	758	*10.479*	851	*20.135*
Malacostraca	O. Isopoda F. Sphaeromatidae *Lekanesphaera hookeri* (Leach 1814)	111	*1.538*	7956	*71.744*	7499	*78.808*	268	*3.349*	29	*0.363*	90	*2.503*	47	*1.022*	1035	*14.833*
	O. Amphipoda F. Corophiidae *Corophium orientale* (Schllenberg 1928)	420	*0.858*	425	*0.602*	796	*1.237*	50	*0.038*	177	*0.266*	572	*0.833*	382	*0.651*	2029	*2.411*
	O. Decapoda F. Palaemonidae *Palaemonetes varians* (Leach 1814)	1163	*16.102*	2503	*442.170*	651	*42.839*	885	*265.334*	1742	*50.863*	961	*347.709*	441	*26.742*	2538	*202.115*
Euentomata	O. Coleoptera F. Hydrophilidae *Berosus hispanicus* (Küster 1847) *Berosus affinis* (Brullé 1835) *Enochrus bicolour* (Fabricius 1792) F. Hydraenidae *Ochtebiussp* F. Dytiscidae *Hygrotus sp.*	2	*0.008*	154	*10.805*	62	*2.188*	0	*0*	0	*0*	0	*0*	0	*0*	7	*0.051*
	O. Hemiptera F. Corixidae *Sigara stagnalis* (Leach 1817)	1507	*10.658*	5454	*49.425*	2030	*12.675*	377	*3.508*	276	*3.906*	118	*2.290*	158	*2.279*	10266	*66.153*
	O. Odanata F. Coenigrionidae *Ischnura graellsi* (Rambur 1842)	0	*0*	654	*3.496*	356	*8.025*	1	*0.002*	0	*0*	2	*0.070*	0	*0*	17	*0.478*
	O. Diptera F. Chironomidae Tr. Chironomini	351	*0.398*	299	*0.452*	432	*0.650*	1245	*1.556*	758	*0.986*	177	*0.650*	111	*0.365*	2035	*3.685*
	F. Ephydridae *Ephydra sp.*	0	*0*	3	*0.002*	150	*2.115*	0	*0*	0	*0*	0	*0*	0	*0*	0	*0*

Table 8.- Total number (N) and dry mass (B) of each taxonomic group analysed. All samples within each period were pooled together irrespective of treatment. Identification was made with the following keys: Tachet et al (2003), Jansson (1986), Castello (1986), Askew 1988, Ruffo (1989), Ortiz and Jimeno (2001),

Argano (1979), Ghetti and Mc Kenzie (1981) and with the help of specialists (see acknowledgements). See Frisch et al. (in press) for details of relative abundance of different copepod species.

Fig 1.- Aerial counts of waterbirds in Veta la Palma from March 2001 to Jun 2002. Total represent all birds counted in the area, including shorebirds, gulls, etc

Fig 2.- Geometric mean and back-transformed SE of total dry mass, and arithmetic mean and SE of taxa richness and diversity index at the end of each experimental period.

Fig 3.- Geometric mean and back transformed SE of counts of copepods, *Potamopyrgus antipodarum, Cyprideis torosa, Lekanesphaera hookeri, Palaemonetes varians, Corophium orientale, Sigara stagnalis* and chironomid larvae at the end of each experimental period.

Fig 4.- Geometric mean and back-transformed SE of mean individual dry mass (in g) for *Lekanesphaera hookeri, Palaemonetes varians, Corophium orientale, Sigara stagnalis* and chironomids larvae, and arithmetic mean and SE of *Potamopyrgus antipodarum* mass at the end of each experimental period.

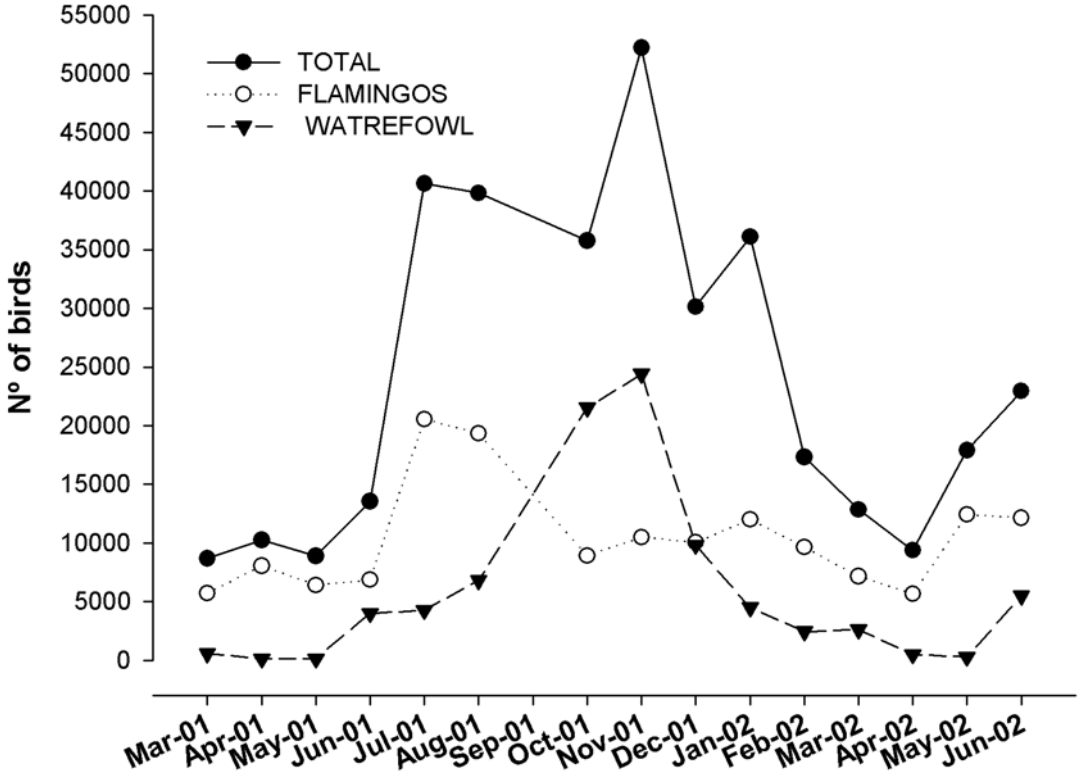

55000
50000
45000
40000
35000
30000
25000
20000
15000
10000
5000
0
Nº of birds
TOTAL
FLAMINGOS
WATREFOWL
Mar-01
Apr-01
May-01
Jun-01
Jul-01
Aug-01
Sep-01
Oct-01
Nov-01
Dec-01
Jan-02
Feb-02
Mar-02
Apr-02
May-02
Jun-02

Fig 1.-

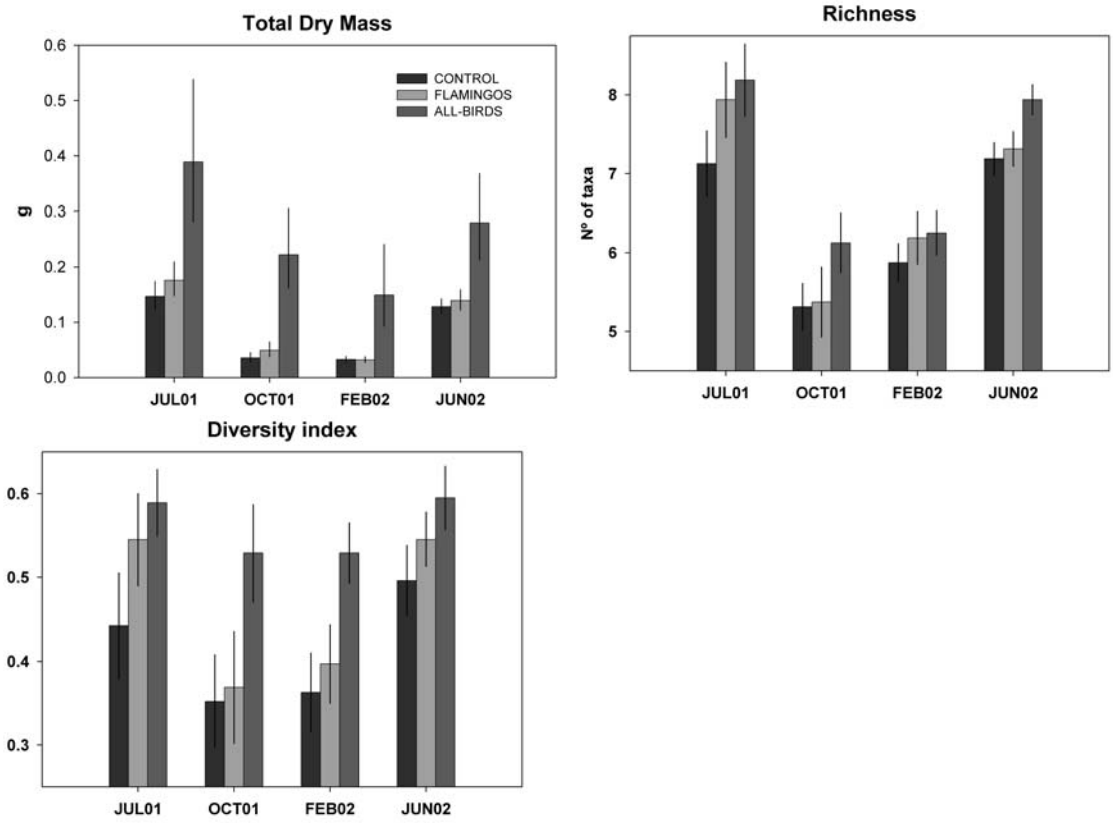

Total Dry Mass
g
0.6
0.5
0.4
0.3
0.2
0.1
0.0
CONTROL
FLAMINGOS
ALL-BIRDS
JUL01
OCT01
FEB02
JUN02
Richness
N° of taxa
8
7
6
5
JUL01
OCT01
FEB02
JUN02
Diversity index
0.6
0.5
0.4
0.3
JUL01
OCT01
FEB02
JUN02

Fig.- 2

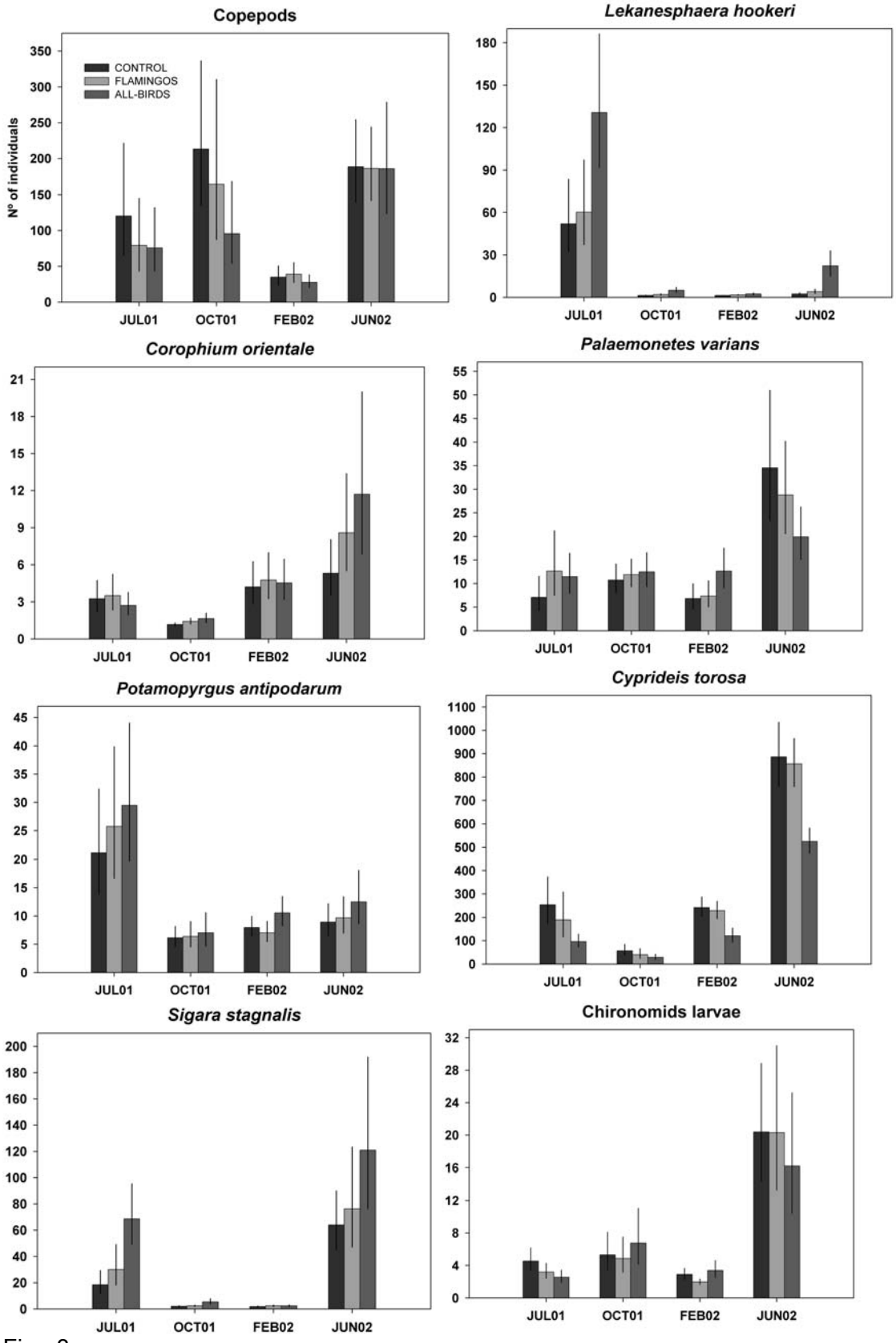

Copepods
CONTROL
FLAMINGOS
ALL-BIRDS
Nº of individuals
JUL01
OCT01
FEB02
JUN02
Lekanesphaera hookeri
Corophium orientale
Palaemonetes varians
Potamopyrgus antipodarum
Cyprideis torosa
Sigara stagnalis
Chironomids larvae

Fig.- 3

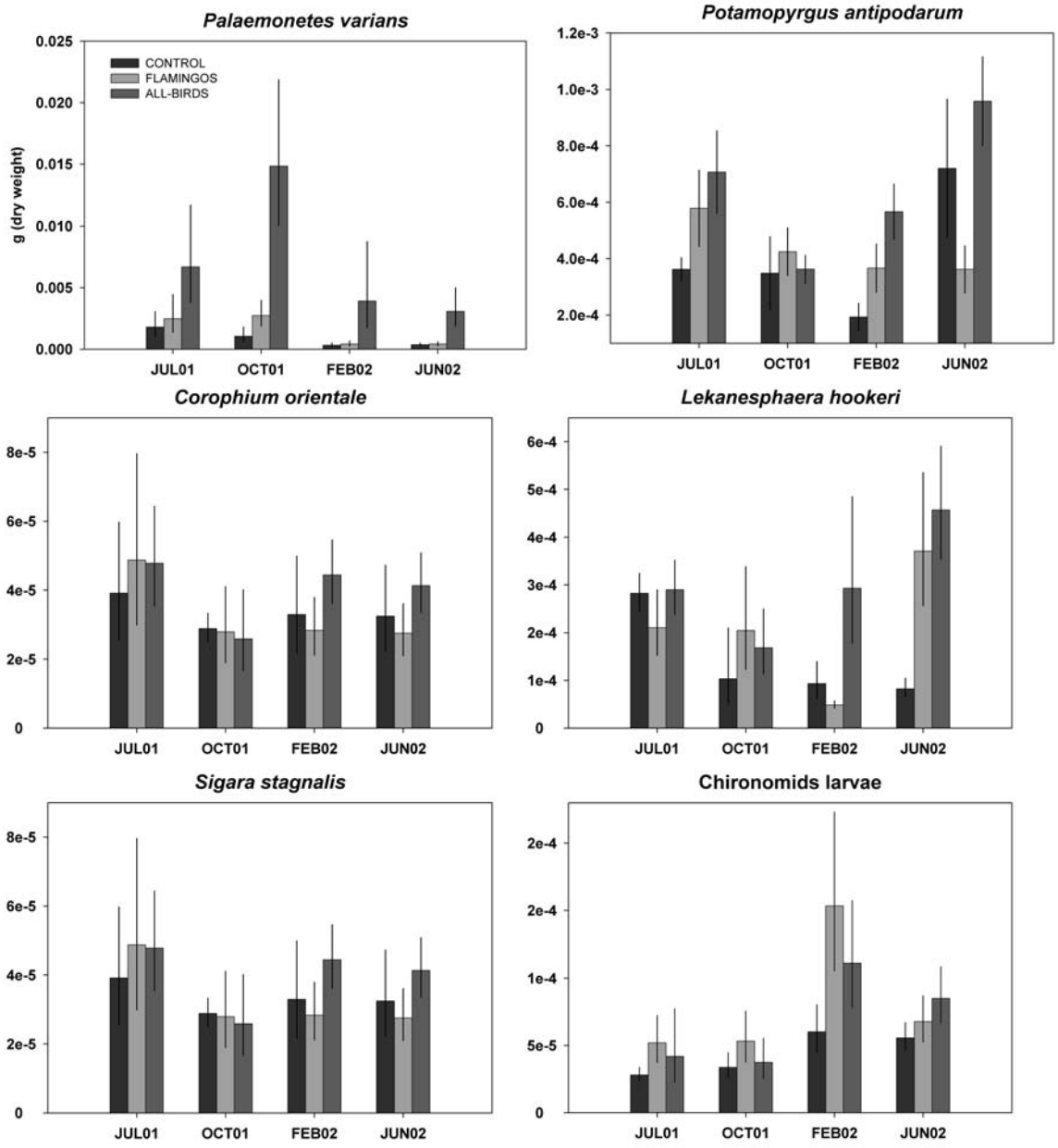

Palaemonetes varians
CONTROL
FLAMINGOS
ALL-BIRDS
g (dry weight)
0.025
0.020
0.015
0.010
0.005
0.000
JUL01
OCT01
FEB02
JUN02
Potamopyrgus antipodarum
1.2e-3
1.0e-3
8.0e-4
6.0e-4
4.0e-4
2.0e-4
Corophium orientale
8e-5
6e-5
4e-5
2e-5
0
Lekanesphaera hookeri
6e-4
5e-4
4e-4
3e-4
2e-4
1e-4
Sigara stagnalis
Chironomids larvae
5e-5

Fig.- 4

CAPÍTULO 4

Efectos ecológicos del flamenco común (*Phoenicopterus ruber*) en la marisma del Parque Nacional de Doñana / Ecological effects of Greater flamingo (*Phoenicopterus ruber*) on the marshland of National Park of Doñana.

Basado en el manuscrito original: Rodríguez-Pérez, H., Green, A.J., and Figuerola, J. Ecological effects of Greater flamingo *Phoenicopterus ruber* in natural marshes in Doñana, SW Spain.

Resumen

Existen sugerencias previas de que los flamencos comunes (*Phoenicopterus ruber*) destruyen los macrófitos, y facilitan el cambio del estado de aguas claras a aguas turbias en las marismas temporales del Parque Nacional de Doñana. Desde el mes de febrero hasta el mes de julio de 2004, se excluyó a los flamencos en veinte cercados de 4x4 metros, distribuidos en dos lucios de la marisma. En julio, se midió los sólidos en suspensión (TSS), la biomasa de macrófitos y la abundancia y el tamaño de los quironómidos bentónicos en las exclusiones y en los controles adyacentes. Hubo menos sólidos en suspensión y más abundancia de macrófitos en el interior de los cercados, pero la diferencia con los controles no fue significativa. Sin embargo, los flamencos causaron una reducción significativa en la abundancia de quironómidos y un incremento en la proporción de larvas más grandes. El efecto de la exclusión fue mayor en el lucio que mantuvo una mayor densidad de flamencos. La duración de la inundación durante 2004 fue especialmente larga, debido a las intensas lluvias, y es posible que los flamencos tengan efectos más fuertes en años con menores precipitaciones.

Palabras clave: flamencos, macrófitos, turbidez, nutrientes, quironómidos, resuspensión del sedimento.

Abstract

It has previously been suggested that Greater flamingos (*Phoenicopterus ruber*) destroy submerged macrophytes and promote a switch from a clear water to a turbid water state in the temporary marshes of Doñana National Park. We excluded flamingos from twenty 4x4 m plots distributed between two lucios (shallow seasonal lakes) within the marshes from February to July 2004. In July, we measured total suspended solids (TSS), macrophyte biomass and the abundance and size of benthic chironomid larvae in exclosures and adjacent control plots. TSS was lower and macrophyte biomass was higher in exclosures, but not significantly so. However, flamingos were found to cause a significant reduction in chironomid abundance and an increase in the proportion of larger larvae. The effect of exclusion was greater in the lucio where the density of flamingos was highest. The duration and extent of flooding were especially high in 2004 owing to heavy rains, and flamingos may have stronger effects in years of lower rainfall.

Keywords: Flamingos, macrophytes, turbidity, nutrients, chironomids, sediment resuspension.

INTRODUCTION

Submerged vegetation has a major functional role in shallow wetlands, e.g. because they provide refuge for invertebrates, change the nutrient dynamics of the system, and prevent resuspension of the sediments (Jeppesen et al 1998, Scheffer 1998). It is therefore important to establish the effects of birds and other biota on the presence and abundance of submerged vegetation. The role of herbivorous waterbirds such as swans, geese and coots has been studied extensively, and is an important factor in the restoration of shallow lakes (Van Donk et al. 1994, Van Donk and Otte 1996, Perrow et al. 1997, Sondergaard et al. 1996). It is widely assumed that the disturbance of sediments by benthivorous fishes can inhibit the colonization of submerged plants and enhance sediment resuspension (Scheffer et al. 1993, Scheffer 1998). However, the potential effects of benthivorous birds on submerged plants have largely been ignored.

It has previously been suggested that Greater flamingos (*Phoenicopterus ruber*) have negative effects on submerged plants and on herbivorous birds in the marshes of Doñana National Park in Spain (Duarte et al. 1990, Grillas et al. 1993, Montes and Bernués 1991) and in the Camargue in France (Gallet 1950). Montes and Bernués (1991) compared areas in the marshes of Doñana National Park with and without concentrations of flamingos, and found their presence to be associated with a decrease in macrophyte biomass and an increase in turbidity and in phytoplankton abundance (as measured by chlorophyll a). However, until now there has been no experimental study in the park to test the hypothesis that the action of flamingos is responsible for these patterns. Greater flamingos produce craters of *ca.* 1 m diameter during their trampling feeding behaviour, and are one of the most abundant waterbirds in Mediterranean wetlands with an increasing population size (Wetlands International 2002, Rodríguez-Pérez and Green 2006). They cause damage to rice crops in France and Spain (Tourenq et al. 2001) and have been shown to change sediment properties and to decrease the abundance of benthic invertebrates in Namibia (Glassom and Branch 1997a, Glassom and Branch 1997b). Their footsteps mobilize sediments and nutrients (Comín et al. 1997).

In this study, we carry out an exclosure experiment with the aim of testing the hypothesis that flamingos reduce the biomass of submerged macrophytes and the abundance of benthic invertebrates in temporary marshes of Doñana National Park, whilst increasing turbidity in the water column. We carried out the experiment for over a six month period during a single hydrological cycle, in two areas within the marshes.

Study site

Our study was carried out in 2004 at two "lucios" (El Lobo and Marilópez) located within the 26,000 ha of natural marshes in Doñana National Park (Fig 1, Castroviejo 1993). This park is also protected as a Biosphere Reserve, UNESCO World Heritage site, Ramsar site and an EU Specially Protected Area. "Lucio" is the local name for shallow, seasonal lakes created in depressions within the marsh that remain flooded until the marsh dries up. These temporary natural marshes are subject to great annual variation in the extent and duration of flooding between years in relation to fluctuations in rainfall (Marín & Garcia 2005). At the height of the wet season in winter and early spring, lucios are surrounded by and interconnected via shallower, more temporary areas of marsh (Espinar et al. 2002, Marín & Garcia 2005). The marsh is fed by freshwater (rainfall and runoff) and isolated from tidal influence of the Guadalquivir estuary. The concentration of salts depends on the frequency and the duration of flooding (Serrano et al in press). While drying during the hot and dry summer, the marshland changes from oligohaline to mesohaline (Table 1), with a wide variation of situations depending on distance from freshwater sources, depth, etc (Castroviejo 1993). The National Park and surrounding area is frequented by over 300,000 waterbirds in most winters, with about 25,000 flamingos in winter and about 50,000 in summer (Martí and del Moral 2002, Johnson and Arengo 2005, Aguilera et al 2005).

The 'Lucios' El Lobo and Marilópez are located in the northern part of the Doñana National Park (Fig 1). They have a surface area of *ca*. 120 and 300 ha respectively, and are surrounded by saltmarsh vegetation dominated by perennial *Arthrocnemum*. The maximum depth for both 'lucios' is *ca*. 1 metre, and fluctuates strongly with rainfall and wind direction. The emergent vegetation is dominated by scattered patches of *Scirpus littoralis*. During our study the dominant submerged macrophytes were *Ruppia drepanensis*, *Ranunculus peltatus, Callithriche truncata* and charophytes.

MATERIALS AND METHODS

Experimental plots were established from February 2004, prior to the emergence from the sediment of submerged macrophytes in early spring (Grillas et al. 1993), to July 2004 covering most of the flooding cycle (from November 2003 to July 2004). Two parallel transects of 300-400 metres in length were established in each of the lucio del Lobo (Lobo from hereon) and the lucio de Marilópez (Marilópez from hereon, Fig. 1). The minimum distance between the two transects within each Lucio was 90 metres. Five pairs of flamingo exclosures and control plots were established along each transect. Each exclosure and its control were position on opposing sides of the transect line, and separated by 20 m. The

distance between pairs of plots was 50-80 m (see Fig 1 for further details). The position of each plot was pinpointed via GPS.

Both exclosures and controls were 4 x 4 m squares delimited by four iron poles pushed vertically into the mud in each corner. Exclosures were created by extending a wire around the poles at a height of *ca*. 70 cm above the bottom, copying a design used successfully in nearby fish ponds (Rodríguez-Pérez and Green 2006). This design takes advantage of the much greater height of flamingos, and the height of the wire was selected after trials confirming that other waterbirds pass comfortably underneath the wire and freely enter the exclosures (Rodríguez-Pérez and Green 2006).

Our plots were visited each month, and physico-chemical measurements were taken. The water depth was measured (always at the same corner of each plot), turbidity was measured with an 8 cm Secchi disk and temperature, conductivity and salinity were measured with a WTW multiprobe 340-i device. During visits, we also took spot measurements of turbidity (using a Hanna HI 93703 probe in Formazine Turbidity Units (FTU) equivalent to Nephelometric Turbidity Units) from different areas of the each lucio that were occupied by flamingo concentrations at that time, together with other points without bird concentrations. We counted the flamingos present in each lucio with 8 x 40 binoculars and recorded their behaviour (roosting or feeding).

On our final visit on 7-8 July as the flooding cycle was approaching its end, we took samples of macrophytes, chironomids and total suspended solids (TSS) from each plot. TSS was measured with a gravimetric method (APHA 1999). One litre of water was carefully taken on arrival, being careful to ensure that the sediment cloud caused by our steps could not influence the measurement. Each sample was later resuspended in the lab, then a known volume of sample was filtered through a Whatman GF/C filter which had previously been dried and weighed. Each saturated filter was then dried for 48 h at 70º C, kept in a desiccator until the environmental temperature was reached, and then weighed with a precision balance (to the nearest 0.0001 g).

We gathered four macrophyte samples per exclosure with a PVC pipe section (0.125 m^2 cross-sectional area) that was pushed into the sediments. All water was extracted with a bowl, and then all macrophyte stems and leaves were carefully cut at the base with finger nails and removed. To avoid edge effects, the pipe was inserted 1 m in towards the centre of the plot from the middle of each side of the square. At the lab, samples were rinsed with tap water to clean off mud and invertebrates, and dried at 70ºC for 48 h prior to weighing.

Benthos was sampled by taking four core samples of the 5 cm of sediments with a 5.5 cm diameter corer from each plot. Benthic samples were taken from undisturbed spots away from the edge and close to those sampled for macrophytes. At the lab, samples were stored in a refrigerator until they could be filtered through a 0.5 mm sieve. Only chironomid larvae

were retrieved and preserved in formalin. We restricted the analysis to this group as the dominant benthic organism in our samples, and because it is so important as prey for waterbirds (del Hoyo et al 1992). Chironomid larvae were identified to tribe level. The length of these larvae were later measured under a binocular microscope with the aid of a digital image system to the nearest 0.01 µm. Prior to statistical analysis, the data from four samples from each plot were pooled, since they were not spatially independent.

Furthermore, we used the aerial monthly census of waterbirds carried out in Doñana National Park and its surroundings. We cite total numbers counted for the whole census of the park and adjacent areas (including the Veta la Palma fish ponds, see discussion).

Statistical analysis

We used generalized mixed linear models (GLMs, McCullagh and Nelder 1989) to analyse the effect of treatment (flamingo exclosure and control), site (Lobo and Marilópez) and water depth. Treatment and site were included as fixed factors, and depth as a continuous variable. The transect and exclosure/control pair nested within transect were included as random factors in the analysis, using GLIMMIX procedure (SAS Institute 2005). We initially included Treatment x Site interactions but excluded them when they were not significant ($p>0.05$). When *post-hoc* analysis of least-squared means was performed, the α significance level ($\alpha=0.05$) was adjusted with False Discovery Rate (FDR) techniques for repeated-tests (Garcia 2003, Garcia 2004).

For TSS and macrophyte biomass, we selected those error distributions and link functions that prevented heteroscedasticity, and deviation from model assumptions. TSS was analysed with an identity link function and lognormal error distribution. Macrophyte biomass was $\log_{10}$ transformed and modelled with an identity link function and normal error distribution. For chironomid counts, we used a log link function and negative binomial error distribution (Gray 2005).

A logistic regression (Crawley 1993) with a logit link and binomial error distribution was used to analyze the size distribution of chironomids larvae. The dependent variable was the proportion of chironomid larvae that were smaller or equal to the median size observed (6.475 mm) in control plots. The numerator of the dependent variable was the number of larvae ≤ 6.475 mm, and the denominator was the total number of larvae.

Graphs of TSS, macrophyte biomass and chironomid abundance were produced using geometric means so as to better represent the skewed data distributions. These were calculated by $\log_{10}$ transformation of the raw data (adding 1 for the number of chironomids, owing to the presence of zeros), calculation of the arithmetic mean and SE, followed by back-transformation.

RESULTS

During the course of the experiment, a steady increase in evaporation together with a lack of water input produced a gradual decline in depth (Table 1), followed by desiccation by the end of July. Sharp rises in temperature, salinity and conductivity occurred over time, and the lucios changed from oligohaline to mesohaline (Table 1).

Marilópez held more flamingos throughout the study than Lobo, and had a higher density of flamingos from February to June inclusive (Fig 2). The numbers of flamingos using both lucios increased towards the end of the experiment as shallower areas of marsh in Doñana dried out, and as the total numbers of flamingos in Doñana increased (Fig. 2). Throughout the study, flamingos were observed using the areas where our transects were placed. However, until June, most of the flamingos were roosting in flocks, whereas from June onwards they spread out more across the lucios and increased feeding activity. Turbidity was visibly increased at the points where flamingos were feeding. Thus, on 18 June turbidity spot measurements in points of Lobo not disturbed by flamingos were 20.46 and 36.7 FTU, whereas in points disturbed by flamingos they were 105 and 172 FTU. Similar measurements for Marilópez were 18.35 and 26.4 for points without flamingos, and 87 and 121 FTU in points with active flamingos. The lowest turbidity values (2.2 FTU in Lobo and 2.3 in Marilópez) were recorded in May, when the densest mats of macrophytes occurred.

In a GLM analyzing TSS data collected from experimental plots in July, there were no significant effects of treatment (control or exclosure), site (lucios) or water depth (Table 2). However, TSS tended to be higher in Marilópez and in flamingo exclosures (Fig. 3).

At the time of sampling, *Ruppia drepanensis* was the only submerged macrophyte still in flower, the other species (see study area) already having ended their reproductive cycles. *R. drepanensis* represented the great majority of the biomass of submerged macrophytes sampled. Macrophyte biomass did not differ significantly between treatments or with depth, but there was a highly significant effect of site, with more biomass in Marilópez (Table 2).

Both tribes Chironomini and Tanytarsini were represented amongst chironomid larvae. A highly significant site main effect in a GLM of larval abundance showed that the density of larvae was highest in Marilópez (Table 3, Fig. 3). The effect of treatment varied between lucios, as shown by a highly significant Treatment x Site interaction (Table 3). Post-hoc tests showed larval abundance to be significantly higher in exclosures than controls for Marilópez, with no treatment effect for Lobo (control vs. exclosure: Marilópez t_{27}=-2.43, p= 0.032; Lobo t_{27}=0.42, p= 0.2; Fig. 3).

In a GLM of larval size, treatment had a significant effect in the absence of a site effect (Table 3). The treatment x site interaction was not significant (p = 0.9). A higher proportion of

large larvae were found in controls than in exclosures (Fig. 4). Whereas 58% of larvae in exclosures were less than 6.475 mm in length, this was true for only 50% of those in controls (Fig). Water depth did not have a significant effect in any of the chironomid analyses.

DISCUSSION

Our exclosure experiment did not provide statistical support for previous suggestions (Montes and Bernués 1991, Duarte et al. 1990, Grillas et al. 1993) that greater flamingos reduce biomass of submerged macrophytes and increase turbidity in the natural marshes of Doñana National Park. Direct observation shows that feeding flamingos do damage macrophytes and create clouds of suspended sediments in the precise points where they are standing, as indicated by our spot turbidity measurements. However, our results indicate that these effects were not detectable at a broader scale across a lucio after five months of exclusion. Montes and Bernués (1991) carried out an observational study in 1986-88 in which they showed that areas without flamingos had more macrophytes and a lower turbidity. These results are not necessarily the product of a causal relationship, and could alternatively be a consequence of flamingos showing a preference for areas with less macrophytes (e.g. because it may facilitate their feeding on benthos). However, despite our results, there are several lines of evidence that suggest that flamingos can have important impacts on macrophytes in Doñana.

Firstly, owing to poor visibility we were unable to quantify macrophyte cover in a non-destructive manner during the course of our study, and it is possible that significant effects may have occurred earlier (e.g. biomass may have been faster to reach a peak in exclosures). Secondly, the strength of flamingo effects on macrophytes are likely to be highly variable in space and time, and it is possible we would have detected strong effects in other parts of the marshes or in other years. A significant effect of flamingos on *Ruppia maritima* has already been recorded in extensive fish ponds in Veta la Palma, an area adjacent to Doñana National Park (Rodríguez-Pérez and Green 2006). Since most of these ponds were created in 1993, the density of flamingos in the National Park has decreased, and Veta la Palma has become the preferred area for flamingos (Aguilera et al. 2004), which are the dominant waterbird species there in terms of biomass (Rodríguez-Pérez and Green 2006). Our results show that a stronger effect of flamingos on chironomid abundance was observed at the site with a higher density of flamingos (Marilópez). Such density effects are also to be expected for macrophyte impacts, and we observed a clear trend for lower TSS in exclosures in Marilópez (Fig. 3).

Thus, the impact of flamingos on macrophytes in the natural marshes may have been reduced since the Veta la Palma fish ponds were created. Our study year (2004) was very

wet with a particularly high extension and biomass of submerged macrophytes across the temporary marshes of the National Park. Stronger impacts in the fish ponds than natural marshes may also be partly due to the higher salinity and turbidity in the ponds, which increases the stress on *Ruppia* (Verhoeven 1975) and may increase its susceptibility to other stressors such as waterbirds.

Another potential reason for our failure to detect significant results is the limited size of our exclosures. Flamingos feeding close to our exclosures are likely to have increased turbidity within them, and it is possible we would have had significant results with larger exclosures.

We found flamingos to have a significant effect on benthic chironomid larvae, reducing their density (in Marilópez) and changing their size distribution. Greater flamingos consume chironomid larvae (Johnson 1997), and we have also found them to reduce the density of benthic chironomids and polychaetes in the Veta la Palma fish ponds (Rodríguez-Pérez and Green unpublished a). Exclosure experiments by others have previously shown greater flamingos to have strong effects on other benthic invertebrates (Glassom and Branch 1997 a,b), and Andean flamingos (*Phoenicoparrus andinus*) to have major effects on benthic fauna and flora (Hurlbert and Chang 1983).

The exclosure effects we observed on chironomids are clearly attributable to flamingos. The only other birds in the area with sufficient height to be excluded were small numbers of Glossy Ibis (*Plegadis flacinelus*), but none was seen near exclosures and this species preys mostly on beetles and Odonata larvae (Macías et al 2004). Although we only found chironomid abundance to be higher in exclosures in Marilópez, where flamingo density was higher, this does not necessarily indicate that flamingos were not consuming chironomids in Lobo as well. Other experiments excluding waterbirds known to feed on chironomids have not always produced detectable effects (Smith et al. 1986, Ashley et al. 2000).

We also found chironomid larvae to be larger in controls. If flamingos were size selective predators of chironomid larvae, we would expect the opposite effect, i.e. larvae to be smaller in controls (Sánchez and Green 2006). As flamingos have fine lamellae capable of filtering plankton (Zweers et al. 1995), it seems unlikely they would select larger larvae. One possible explanation for our size effect is that an increase in larval density in exclosures led to a reduced size due to inhibition of growth rates by competition (Armitage et al. 1995). However, this could not explain why we recorded an interaction between site and treatment for larval density but not for size. Another explanation could be a difference between treatments in the relative abundance of different chironomid species of different size (see Fuentes et al. 2005). It is noteworthy that, although controls had a higher proportion of larvae with a length > 6.475 mm, the maximum length was recorded in exclosures (Fig. 4).

The exclusion of vertebrate predators often leads to indirect effects on the size distribution of benthic invertebrates, mediated via competition or interactions with invertebrate predators themselves released from predation pressure ([Sih, 1985 #275] [Thrush, 1999 #254]). It is possible that the density of predatory invertebrates, such as beetles or dragonfly larvae, increased in flamingo exclosures leading to a reduction in chironomid size. However, such predatory invertebrates may not be selective amongst the size range of chironomid larvae we recorded (Wellborn et al. 1996). Results of an exclosure study in the Veta la Palma fish ponds (Rodríguez-Pérez and Green unpublished a, b) illustrates the complexities of the relationship between flamingos and chironomid larvae. The density of larvae in sediments was higher in flamingo exclosures than in controls, but the size (quantified as mean mass) was no different. In contrast, there was no effect of treatment on the density of larvae collected from the water column, but the size was higher in exclosures (i.e. the opposite effect to that recorded in the present study).

Conservation management has led to a marked increase in the size of the Greater flamingo population in Doñana and across the Mediterranean region since 1970, making this one of the most abundant breeding waterbirds in terms of biomass (Johnson 1997, Wetlands International 2002, Aguilera et al 2004). Although there was a decrease in the numbers of flamingos using the natural marshes of Doñana following the creation of fish ponds, numbers there have began to recover in recent years (Aguilera et al 2004) with a record 16,000 pairs nesting there in 2003 (Johnson and arengo 2004). As the whole Spanish population continues to increase, numbers in the National Park are also likely to continue increasing. Thus, the chance of flamingos reaching sufficient densities to have an important impact on macrophytes and benthos may increase in the future, especially in years of low rainfall when flamingos are more concentrated in available habitat than in 2004.

In conclusion, greater flamingos have a pronounced effect on the benthic invertebrates in temporary marshes, as has previously been observed in more permanent habitats. Although they reduce the cover of submerged vegetation in brackish fish ponds, this has yet to be demonstrated conclusively for natural marshes. More research is required to understand the nature of spatial and temporal variation in the relationship between flamingos and submerged vegetation.

References:

American Public Health Association. 1999.Standard Methods for the examination of water and wastewater. APHA.

Ashley, M. C., J. A. Robinson, L. W. Oring, and G. A. Vinyard. 2000. Dipteran standing stock biomass and effects of aquatic bird predation at a constructed wetland. Wetlands 20:84-90.

Bulthuis, D. A., G. W. Brand, and M. C. Mobley. 1984. Suspended Sediments and Nutrients in Water Ebbing from Seagrass-Covered and Denuded Tidal Mudflats in a Southern-Australian Embayment. Aquatic Botany 20:257-266.

Castroviejo, J. 1993. Mapa del Parque Nacional de Doñana; Memoria. Consejo Superior de Investigaciones Científicas.

Comín, F. A., J. A. Herrera-Silveira, and M. Martín. 1997. Flamingo footsteps enhance nutrients release from the sediment to the water column. Wetlands International Publications. Limnology and waterfowls. Monitoring, modelling and management. Proceedings of a Symposium on Limnology and Waterfowl. 43:211-227.

Del Hoyo, J., Elliot, A. and Sargatal, J. (eds.). 1992. Handbook of the birds of the world. - Lynx Editions.

Duarte, C. M., C. Montes, S. Agustí, P. Martino, M. Bernués, and JC. Kalff. 1990. Biomasa de macrófitos acuáticos en la marisma del Parque Nacional de Doñana (SW España); Importancia y factores ambientales que controlan su distribución. Limnetica 6:1-12.

Espinar, J. L., García, L. V., García Murillo, P. & Toja, J. 2002. Submerged macrophyte zonation in a Mediterranean salt marsh: a facilitation effect from established helophytes? *Journal of Vegetation Science*, 13, 831-840.

Faafeng, B.A., and M. Mjelde. 1998. Clear and turbid water in shallow norwegian lakes related to submerged vegetation. Pages 361-368 in Jeppensen, E., M. Sondergaard, M. Sondergaard, and K. E. Christoffersen. 1998. The structuring role of submerged macrophytes in lakes. Spring-Verlag, New York.

García-Novo, F. & Marín, C. 2005. Doñana, Agua y Biosfera. Confederación Hidrográfica del Guadalquivir, Ministerio de Medio Ambiente. http://www.chguadalquivir.es/opencms/opencms/chg-web/agua_y_biosfera.html

Garcia, L. V. 2003. Controlling the false discovery rate in ecological research. Trends in Ecology & Evolution 18:553-554.

Garcia, L. V. 2004. Escaping the Bonferroni iron claw in ecological studies. Oikos 105:657-663.

Glassom, D., and G. M. Branch. 1997a. Impact of predation by greater flamingos *Phoenicopterus ruber* on the macrofauna of two southern African lagoons. Marine Ecology Progress Series 149:1-12.

Glassom, D., and G. M. Branch. 1997b. Impact of predation by greater flamingos *Phoenicopterus ruber* on the meiofauna, microflora, and sediment properties of two southern African lagoons. Marine Ecology Progress Series 150:1-10.

Gray, B. R. 2005. Selecting a distributional assumption for modelling relative densities of benthic macroinvertebrates. Ecological Modelling 185:1-12.

Grillas, P., P. Garcia-Murillo, O. Geertz-Hansen, N. Marbá, C. Montes, C. M. Duarte, L. Tan Ham, and A. Grossman. 1993. Submerged macrophyte seed bank in a Mediterranean temporary marsh: abundance and relationship with established vegetation. Oecologia 94:1-6.

Hargeby, A., G. Andersson, I. Blindow, and S. Johansson. 1994. Trophic Web Structure in a Shallow Eutrophic Lake During a Dominance Shift from Phytoplankton to Submerged Macrophytes. Hydrobiologia 280:83-90.

Hoyer, M. V., and D. E. Canfield. 1994. Bird Abundance and Species Richness on Florida Lakes - Influence of Trophic Status, Lake Morphology, and Aquatic Macrophytes. Hydrobiologia 280:107-119.

Hurlbert, S. H., and C. C. Y. Chang. 1983. Ornitholimnology - Effects of Grazing by the Andean Flamingo (Phoenicoparrus-Andinus). Proceedings of the National Academy of Sciences of the United States of America-Biological Sciences 80:4766-4769.

Jeppensen, E., M. Sondergaard, M. Sondergaard, and K. E. Christoffersen. 1998. The structuring role of submerged macrophytes in lakes. Springer-Verlag, New York.

Jeppesen, E., M. Sondergaard, J.P. Jensen, E. Kanstrup and B. Petersen. 1998. Macrophytes and turbidity in brackish lakes with special emphasis on the role of top-dwon control. Pages 369-380 in Jeppensen, E., M. Sondergaard, M. Sondergaard, and K. E. Christoffersen. 1998. The structuring role of submerged macrophytes in lakes. Spring-Verlag, New York.

Johnson, A. R. 1997. Long-term studies and conservation of Greater Flamingos in the Camargue and Mediterranean. Colonial Waterbirds 20:306-315.

Johnson and Arengo (Coord.) 2005 Flamingo specialist group. Newsletter nº 12. Annual reports 2002-2004. Wetland International

Macías, M., A.J. Green, and M.I. Sánchez. 2004. The diet of Glossy Ibis *Plegadis falcinellus* in Doñana, south west Spain. Waterbirds 27:234-239.

Mc Cullagh, P. and Nelder, J.A. 1989. Generalized linear models. Chapman and Hall.

Montes, C., and M. Bernués. 1991. Incidencia del Flamenco Rosa (Phoenicopterus ruber roseus) en el funcionamiento de la marisma del Parque Nacional de Doñana (SW

España). Pages105-110 in Pintos, M.R., S. Prieto, M. Redón y A. Johnsson . 1991. REunión técnica sobre la situación y problemática del flamenco rosa (*Phoenicopterus ruber roseus*) en el Mediterráneo Occidental y África Noroccidental. Agencia de Medio Ambiente de la Junta de Andalucía.

Noordhuis, R., D. T. van der Molen, and M. S. van den Berg. 2002. Response of herbivorous water-birds to the return of Chara in Lake Veluwemeer, the Netherlands. Aquatic Botany 72:349-367.

Perrow, M. R., J. H. Schutten, J. R. Howes, T. Holzer, F. J. Madgwick, and A. J. D. Jowitt. 1997. Interactions between coot (Fulica atra) and submerged macrophytes: the role of birds in the restoration process. Hydrobiologia 342/343:241-255.

Rodríguez-Pérez, H., and A.J. Green. 2006. Waterbird impacts on widgeongrass Ruppia maritime in a Mediterranean wetland: comparing bird groups and seasonal effects. Oikos 112: 525-534.

SAS Institute Inc. 2005. SAS GLIMMIX.

Scheffer, M., S. H. Hosper, M.-L. Meijer, B. Moss, and E. Jeppesen. 1993. Alternative equilibria in shallow lakes. Trends in Ecology and Evolution 8:275-279.

Scheffer, M. 1998. Ecology of shallow lakes. 357 pp. Kluwer Academic Publisher, Dordrecht.

Serrano, L., M. Reina, G. Martin, I. Reyes, A. Arechenderra, D. León and J. Toja. In press. The aquatic systems of Doñana (SW Spain): watersheds and frontiers. Limnetica.

Smith, L. M., L. D. Vangilder, R. T. Hoppe, S. J. Morreale, and I. L. Brisbin. 1986. Effect of diving ducks on benthic food resources during winter in South Carolina, USA. Wildfowl 37:136-141.

Sondergaar, M., B. L., T. Lauridsen, E. Jeppesen, and T. Vindbaek Madsen. 1996. The impact of grazing waterfowl on submerged macrophytes: In situ experiments in ashallow eutrophic lake. Aquatic Botany 53:73-84.

Sondergaard, M., P. Kristensen, and E. Jeppesen. 1992. Phosphorus release from resuspended sediment in the shallow and wind-exposed Lake Arreso, Denmark. Hydrobiologia 228:91-99.

Thrush, S. F., R. D. Pridmore, J. E. Hewitt, and V. J. Cummings. 1994. The Importance of Predators on a Sandflat - Interplay between Seasonal-Changes in Prey Densities and Predator Effects. Marine Ecology-Progress Series 107:211-222.

Tourenq, C., Aulagnier, S., Durieux, L., Lek, S., Mesléard, F., Johnson, A. and Martin, J.L. 2001. Identifying rice fields at risk from damage by greater flamingo. - J. Appl. Ecol. 38:170-179.

Vadeboncoeur, Y., M. J. Vander Zanden, and D. M. Lodge. 2002. Putting the lake back together: Reintegrating benthic pathways into lake food web models. Bioscience 52:44-54.

Van Donk, E., E. De Deckere, J. G. P. Klein Breteler, and J. T. Meulemans. 1994. Herbivory by waterfowl and fish on macrophytes in a biomanipulated lake: effects on long-term recovery. Verhandlungen Internationale vereinigung für teoretische und andwebante limnologie 25:2139-2143.

Van Donk, E., and A. Otte. 1996. Effects of grazing by fish and waterfowl on the biomass and species composition of submerged macrophyes. Hydrobiologia 340:285-290.

Vareschi, E. 1978. The ecology of lake Nakuru (Kenya). Abundance and feeding of lesser flamingo. Oecologia 32:11-35.

Verhoeven, J. T. A. 1975. *Ruppia*- communities in the Camargue, France. Distribution and structure in relation to salinity and salinity fluctuations. *Aquatic Botany*, 1, 217-241.

Wellborn, G. A., D. K. Skelly, and E. E. Werner. 1996. Mechanisms creating community structure across a freshwater habitat gradient. Annual Review of Ecology and Systematics 27:337-363.

Wetlands International. 2002. Waterbird population estimates. - Wetlands International.

Zweers, G., De Jong, F. & Berkhoudt, H. 1995. Filter feeding in Flamingos (*Phoenicopterus ruber*). The Condor, 97, 297-324.

	Marilópez					
	4th February	**10th March**	**23th April**	**11th May**	**18th June**	**7th July**
Temperature ºC	18	18	26	19	28	30
Salinity %	0.7	0.4	0.7	0.9	2.9	6.6
Conductivity mS/cm	1.76	1.21	1.67	2.12	5.3	11.38
Depth Min-Max cm	46-51	63-69	52-57	50-53	25-30	8-16
Secchi Min-Max cm	14-19	24-51	22-Bottom	Bottom	5-17	2-Bottom

	El Lobo					
	4th February	**10th March**	**21th April**	**12th May**	**18th June**	**8th July**
Temperature ºC	18	18	20	22	27	35
Salinity %	0.7	0.4	0.8	1	3.2	6.8
Conductivity mS/cm	1.72	1.15	1.89	2.28	5.98	10.93
Depth Min-Max cm	46-51	61-69	47-53	45-50	25-29	12-18
Secchi Min-Max cm	16-21	14-26	30-Bottom	Bottom	12-Bottom	9-Bottom

Table 1.- Physical and chemical features of lucios Marilópez and El Lobo on sampling dates in 2004. 'Bottom' indicates that the Secchi disk was visible at the bottom, indicating high visibility.

	Total suspended solids						Macrophyte biomass					
	Estimate	SE	df_N	df_D	F	p	Estimate	SE	df_N	df_D	F	p
Intercept	4.18	0.21					-1.03	0.07				
Site			1	28	4.20	**0.05**			1	28	35.59	**<0.0001**
El Lobo	-0.51	0.25					-0.4	0.07				
Treatment			1	28	1.64	0.2			1	28	0.23	0.6
Control	0.32	0.25					-0.04	0.03				

Table 2.- Summary of generalized linear models testing the main factors site (two levels) and treatment (two levels) on total suspended solids (TSS, g/l) and macrophyte dry biomass (g/m^2, log_{10} transformed). The interaction was not significant ($p>0.05$) and was removed from the analysis. Transect and flamingo exclosure/control pair were included as random factors using the GLIMMIX procedure, with identity link function and lognormal error distribution for TSS, and normal error distribution for macrophyte biomass. Marilópez and exclosures were aliased. See methods for details.

	Chironomid abundance						Chironomid size					
	Estimate	SE	df_N	df_D	F	P	Estimate	SE	df_N	df_D	F	P
Intercept	3.7	0.3					0.39	0.17				
Site (S)			1	27	9.23	**0.005**			1	24	0.02	0.9
El Lobo	-1.68	0.4					0.03	0.2				
Treatment (T)			1	27	0.50	0.5			1	24	4.44	**0.05**
Control	-0.99	0.4					-0.36	0.17				
S x T			1	27	7.14	**0.01**						n.s.
El Lobo x Control	1.57	0.59										

Table 3.- Summary of generalized linear models testing the main factors site (two levels) and treatment (two levels) on chironomid larvae abundance and chironomid size. When the interaction was not significant ($p>0.05$) it was removed from the analysis. Transect and flamingo exclosure/control pair were included as random factors using the GLIMMIX procedure, with log link function and negative binomial error distribution for chironomid abundance, and logit link and binomial error distribution for chironomid size. Marilópez and exclosures were aliased. See methods for details.

Fig 1.- Map of the study area showing the location within Spain. A schematic drawing shows an example of a transects with five pairs of control plots and flamingo exclosures. The large image is a clip of Landsat TM image composed with the bands 5, 4, and 3 (RBG) for the area of study at 13th of June 2004, showing flooded areas of the marsh in dark grey, Intensity of the grey scale indicates soil moisture. Two straight parallel lines in each lucio basin indicate the transects along which the exclosures and control were set.

Fig 2.- Monthly aerial counts of flamingos at Marilópez (light grey bars), El Lobo (black bars), and total number of flamingos (straight line) counted for the whole Doñana area during the aerial census for the period from January to July 2004. The left y axis shows the scale for lucio counts and right y axis for the whole of Doñana.

Fig 3.- Geometric mean and back-transformed SE of TSS, macrophyte biomass and chironomid counts for both treatments in the two lucios in July 2004.

Fig 4.- Size frequency distribution of chironomids taken from controls and exclosures, combining data for both 'lucios'. The intervals for length in the x axis are of 0.5 mm. N refers to the total number of chironomids gathered in each treatment.

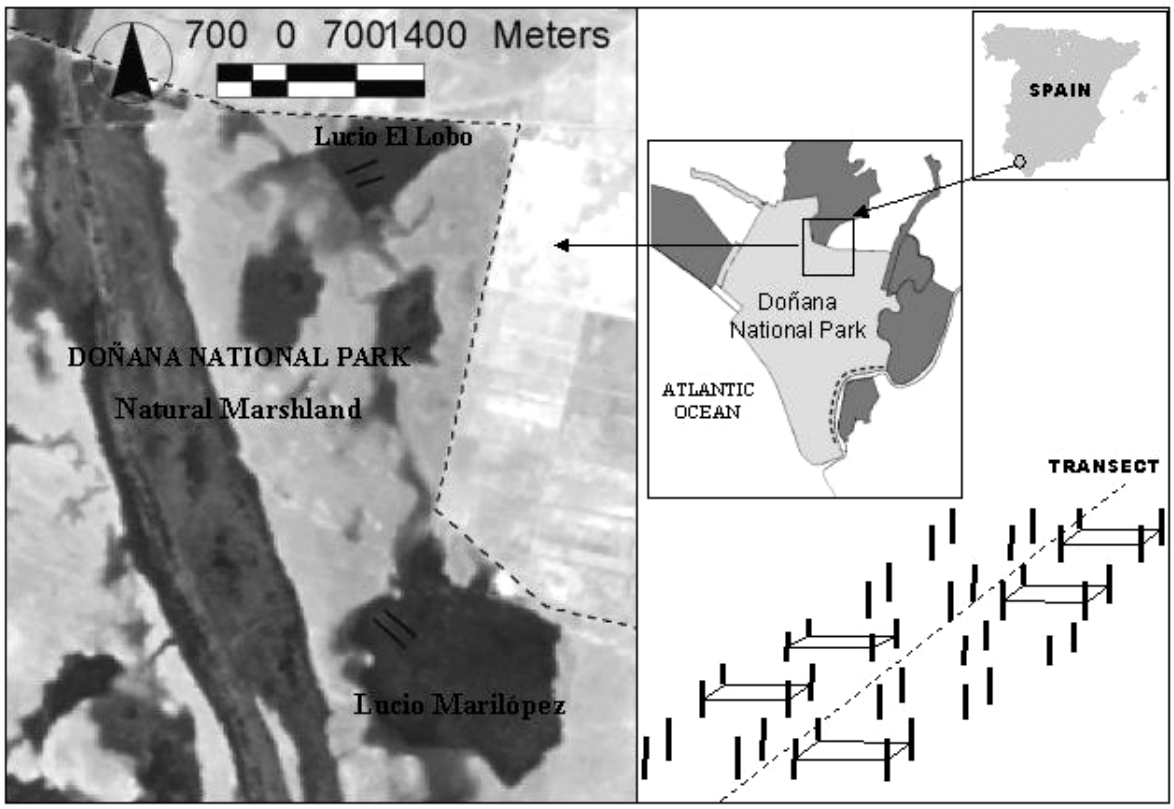

700 0 7001400 Meters
Lucio El Lobo
DOÑANA NATIONAL PARK
Natural Marshland
Lucio Marilópez
SPAIN
Doñana
National Park
ATLANTIC
OCEAN
TRANSECT

Fig 1.-

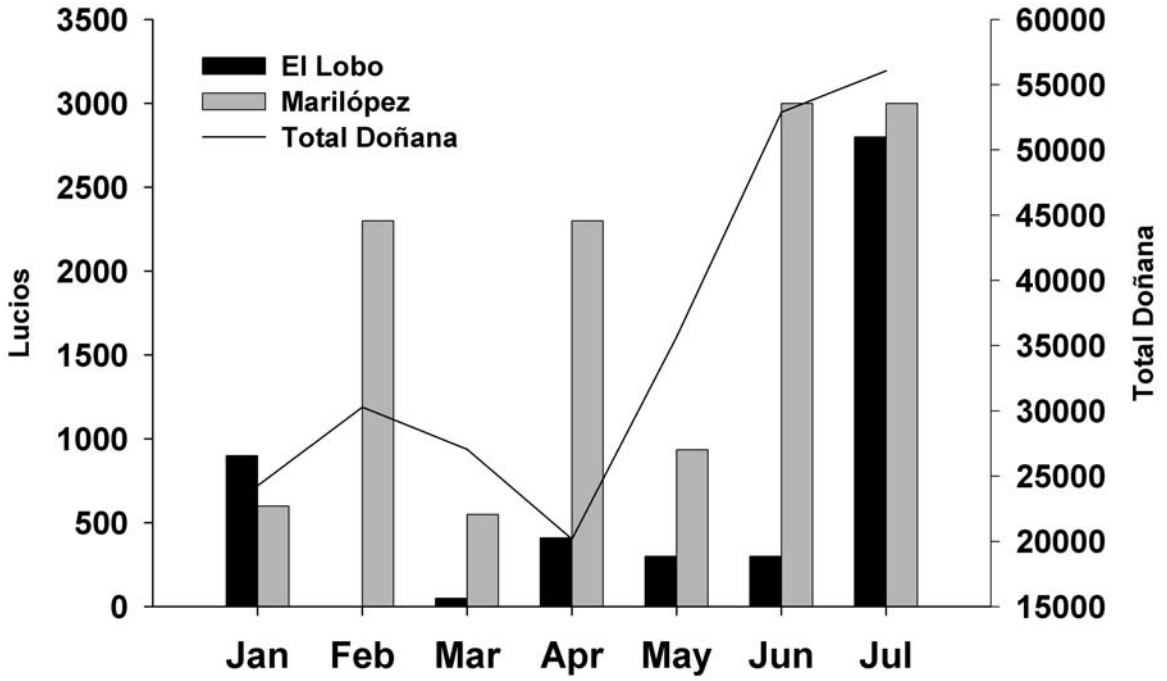
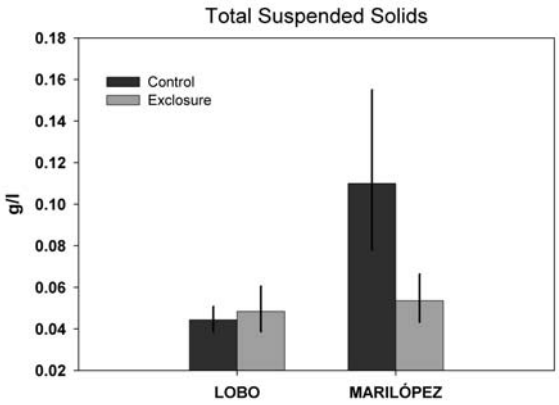
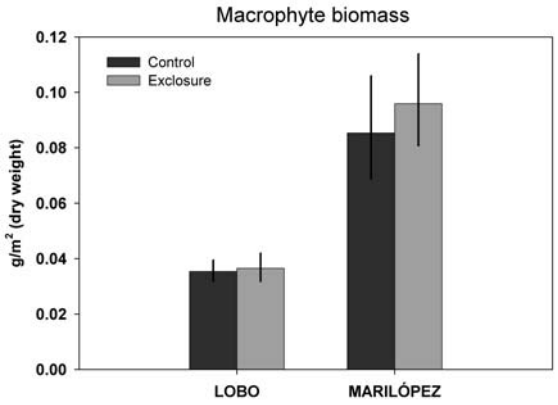

El Lobo
Marilópez
Total Doñana
Lucios
Total Doñana
3500
3000
2500
2000
1500
1000
500
0
60000
55000
50000
45000
40000
35000
30000
25000
20000
15000
Jan
Feb
Mar
Apr
May
Jun
Jul

Fig 2.-


Total Suspended Solids
Control
Exclosure
g/l
0.18
0.16
0.14
0.12
0.10
0.08
0.06
0.04
0.02
LOBO
MARILÓPEZ


Macrophyte biomass
Control
Exclosure
g/m² (dry weight)
0.12
0.10
0.08
0.06
0.04
0.02
0.00
LOBO
MARILÓPEZ

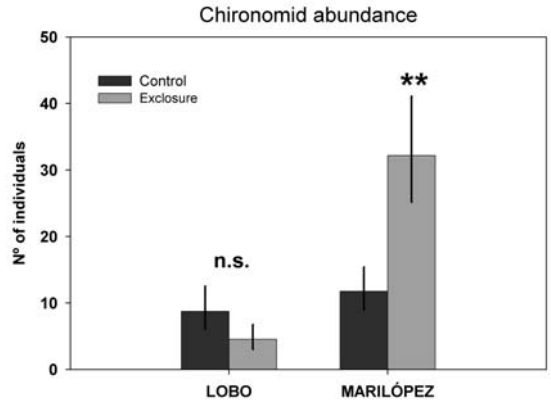

Chironomid abundance
Control
Exclosure
Nº of individuals
50
40
30
20
10
0
**
n.s.
LOBO
MARILÓPEZ

Fig 3.-

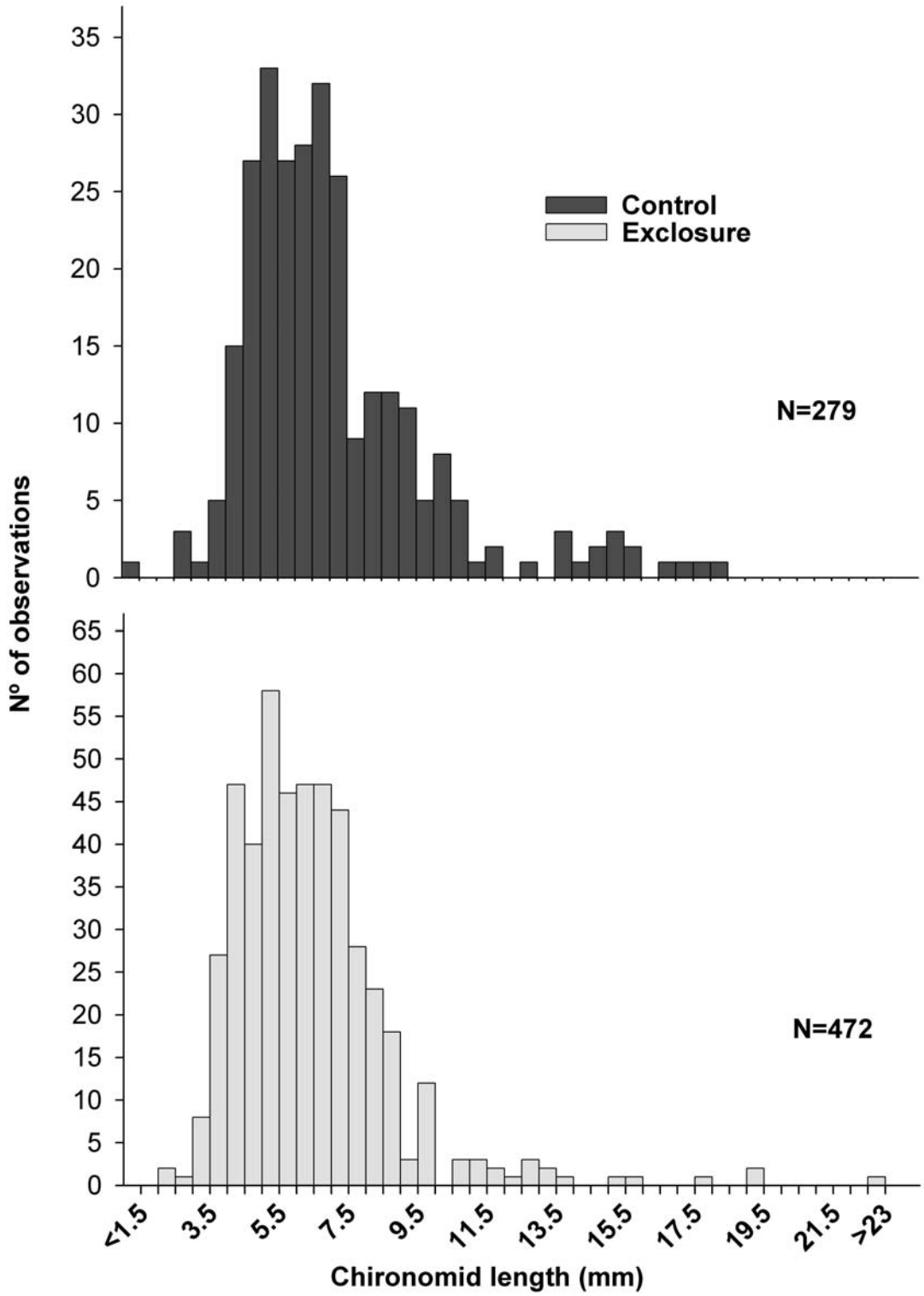

Nº of observations
Control
Exclosure
N=279
N=472
Chironomid length (mm)
<1.5
3.5
5.5
7.5
9.5
11.5
13.5
15.5
17.5
19.5
21.5
>23

Fig 4.-

SÍNTESIS

SÍNTESIS:

Los resultados de esta tesis se han discutido en cada uno de los capítulos de una forma detallada, por lo que en esta sección pretendo ofrecer un visión general de todos el trabajo integrando los resultados de los diferentes capítulos, resaltando los aspectos más importantes, las deficiencias y sugiriendo posibles cuestiones para futuras investigaciones.

Aspectos más destacables.

Hay diversos trabajos sobre la selección de hábitat por distintas especies de aves acuáticas, que han demostrado que, en parte, las aves se distribuyen en función de la abundancia y disponibilidad de su alimento, tanto de plantas acuáticas como de invertebrados. Sin embargo, existen pocos trabajos en la literatura científica que aborden, como en esta tesis, el estudio de los efectos de las aves sobre otros organismos del humedal, integrando tanto efectos en la vegetación sumergida, como en las comunidades de invertebrados. Además son más escasos aún los que comparan los efectos de una única especie, en nuestro caso los flamencos, con los producidos por el conjunto de aves acuáticas al que pertenece dicha especie.

Otro aspecto novedoso de este tesis es haber comprobado los efectos que tienen la aves acuáticas en invertebrados no bentónicos. Y es novedoso hasta tal punto, que en la discusión de los resultados no se han podido comparar con otros trabajos similares, ya que no hemos encontrado ninguno. Los trabajos de este tipo se han centrado, principalmente, en los peces como depredadores de zooplancton o de invertebrados epifíticos.

Este trabajo demuestra de una forma clara que las aves acuáticas juegan un papel importante en los humedales que se han estudiado y es de suponer que efectos similares, de mayor o menor intensidad, se detectarían en otros sistemas. La densidad de aves en Doñana no parece ser especialmente alta comparado con las revisiones anteriores, haciéndonos pensar que los efectos de aves demostrados allí tendrán importancia a escalas mucho más amplias. Los resultados para el caso de los estudios realizados en Veta la Palma muestran claramente, que los efectos de la actividad de las aves se manifiestan sobre los principales componentes del sistema, con un control de tipo '*top-down*' de las redes tróficas. Simultáneamente cabe esperar un control de tipo "*bottom-up*" en el que la distribución de aves en un momento dado dependerá de la distribución de su alimento. Hay que destacar además, que los efectos producidos por las aves se hicieron patentes, a pesar de que las balsas en las que se realizaron los experimentos estaban pobladas con peces, y la actividad depredadora de éstos podía haber enmascarado los resultados atribuibles a las aves.

Sin embargo, no se han encontrado evidencias estadística que confirmen las sugerencias previas sobre el papel de los flamencos en la degradación de las praderas de macrófitos y el incremento de turbidez de la columna de agua, en la marisma del Parque Nacional de Doñana. No obstante, aún sin otorgándoles verosimilitud, las tendencias apuntaban a mayores cantidades de biomasa en los cercados que en los controles.

Los resultados sugieren que los efectos de los flamencos se manifiestan de forma más clara sobre los organismos del bentos, tanto por causa de la depredación como por la 'bioturbación'. Las exclusiones completas de todas las aves acuáticas, produjeron efectos más intensos y más amplios, al incluir reducciones de las densidades de muchos invertebrados de la columna de agua y una reducción significativa de la biomasa de *Ruppia maritima*. Estas diferencias hay que atribuirlas principalmente a las anátidas y a las fochas, algunas de ellas claramente herbívoras, sin embargo hay que considerar que la exclusión de los flamencos también contribuyó a encontrar dichos resultados.

Es muy importante también, comprobar como las reducciones significativas en las abundancias de los invertebrados y los macrófitos, así como en los índices de diversidad y riqueza de invertebrados, se han producido de forma independiente al periodo en el que se realizó el experimento durante el ciclo anual que se estudió. Aunque sí se han detectado diferencias en la intensidad de algunas de estas reducciones, asociadas a cambios en la abundancia de las aves, los invertebrados y las plantas, vinculados a las fluctuaciones estacionales de sus poblaciones. De hecho, en muchos de los trabajos publicados que estudian los efectos de las aves sobre algún grupo de organismos, los experimentos se han realizado en momentos propicios para detectarlos (p.ej. grandes acumulaciones durante los periodos migratorios), y se postula que los efectos sólo son detectables en dichos momentos. Sin duda, la significación y la intensidad de cualquier efecto es dependiente de la intensidad del proceso que lo provoca, y por tanto un aumento por encima de cierta cantidad umbral de la densidad de aves, marcará la diferencia entre que los impactos sean o no importantes para el sistema. Por eso se hace imprescindible considerar la intensidad de los efectos de una determinada población de aves en los humedales en los que habita, cuando se establecen medidas de gestión que favorecen a las aves.

En este sentido, el caso del flamenco común (*Phoenicopterus ruber*) en Doñana puede ser importante. En este trabajo se ha demostrado los efectos importantes de su población en un humedal de Doñana y su carácter como especie clave o ingeniera, y aunque los resultados en la marisma del Parque Nacional de Doñana no han sido concluyentes. Si se considera el aumento de la población de flamencos en el área del Mediterráneo, y el hecho de que estén volviendo a registrase densidades altas en la marisma del P.N. de Doñana, se podría suponer que en un futuro los efectos deletéreos sobre la vegetación sumergida en

ciertas zonas podrían llegar a ser evidente. Además la reducción de la biomasa de los invertebrados sería mas intensa de la que ya se ha detectado.

En otros ejemplos de este tipo de experimentos, que excluyen a los depredadores superiores, es muy común encontrar evidencias de efectos indirectos, habitualmente se hacen patentes a través de organismos que se ven favorecidos en la competencia o en la depredación, y que aumentan su tamaño o su número, sin embargo en nuestro ejemplo tan sólo se ha encontrado el caso de los ostrácodos. Como se apunta en las discusiones de los dos capítulos que tratan este tema, es posible que si los experimentos hubieran durado más se hubiesen encontrado efectos de este tipo. Podría ocurrir también que todos los invertebrados estén sujetos a una depredación igual de intensa y por tanto todos se beneficien de la protección de los cercados, aunque con el tiempo es previsible que los efectos de la competencia y de los organismos depredadores produjesen aumentos de algunas especies en concreto. Este hecho parece indicar que el periodo de aproximadamente tres meses, durante el que estuvieron instalados los cercados, ofrece una buena medida del efecto exclusivo de las aves, aunque sería importante contrastar nuestros resultados con los efectos de exclusión durante más tiempo.

Limitaciones más importantes.

Además de los aspectos positivos que creo que tiene la tesis, existen también limitaciones a la hora de poder interpretar los resultados. Una de las más importante para valorar los efectos sobre la comunidad de invertebrados, especialmente en los experimentos en Veta la Palma, es la falta de información previa sobre esta comunidad y el hecho de desconocer cómo se estructura las comunidades de invertebrados acuáticos presentes, las relaciones tróficas que existen entre ellos y su abundancia y distribución en relación a la de los macrófitos. Conocer todos o algunos de estos aspectos podría haber ayudado a interpretar los resultados.

Un aspecto metodológico que también hubiera facilitado la interpretación de los resultados, hubiera sido emplear los 5 cm superiores del sedimento que quedaba confinado por los *corer* que se utilizaron para muestrear los invertebrados de la columna de agua, para muestrear los invertebrados del bentos, y de esa forma tener un solo método para cuantificar los invertebrados de los sedimentos y de la columna de agua. Esto simplificaría los análisis (por ejemplo, no sería necesario realizar dos análisis para el mismo grupo taxonomico como los ostracodos) y facilitaría una visión global de los efectos sobre los invertebrados. Este aspecto creo que puede ser importante en estudios de este tipo, y además es viable realizarlo si se muestrean humedales someros utilizando un diámetro de *corer* adecuado a las características físicas del sedimento.

También resulta limitante para establecer la causa de los efectos de las aves, el método empleado de cercados de exclusión. Aunque está demostrado que las diferentes especies de aves, presentes en las áreas de estudio, se alimentan de las diferentes especies de invertebrados recogidos en las muestras, no se puede discernir qué importancia relativa tiene en los resultados, cada una de las posibles causas de la reducción de la abundancia de invertebrados y los efectos detectados en las otras variables analizadas. A saber: los efectos directos de la depredación, la 'bioturbación' del sedimento para el caso de los invertebrados del bentos, la eliminación de los macrófitos y asociado a ésta reducción la facilitación del acceso a los peces a sus presas.

Para el caso concreto del experimento de la marisma hubiese sido interesante contar con al menos otro año de datos, dada la gran variabilidad interanual de este ecosistema. En nuestro caso, no fue posible repetir el año siguiente (2005) debido a la fuerte sequía aquel año.

Posibles investigaciones futuras.

Consideramos que esta tesis tiene un carácter bastante pionero y demuestra la necesidad de realizar mucha más investigación sobre "ornitolimnología", es decir el papel funcional de las aves en los ecosistemas acuáticos. Esto es así tanto en ecosistemas mediterráneos como en otras partes del mundo.

En el caso concreto de investigaciones futuras en Veta la Palma, sería muy importante realizar una serie de experimentos más a largo plazo, para darle más oportunidad a los posibles efectos indirectos y así entender mejor la importancia relativa de competencia y prelación en este sistema. Sería especialmente interesante abordar el estudio de la red trófica de las balsas de Veta la Palma, incorporando a todos los componentes del sistema, de forma que se pudiese valorar la intensidad de las interacciones tróficas que existen entre los organismos que participan en la red. Este tipo de estudio hoy en día es abordable con el empleo de técnicas de medición de la proporción de isótopos estables pesados de carbono y de nitrógeno, y complementando esta información con análisis de la dieta de los organismos (análisis de tractos digestivos, heces y egagrópilas). Los resultados de este tipo de trabajo, ayudarían a establecer la importancia de la depredación de las aves frente a las otras posibles causas, argumentadas en el apartado anterior, en los resultados de esta tesis. Por supuesto esto es generalizable a otros humedales.

En este sentido esclarecer la importancia la alteración del hábitat producida por las aves, 'bioturbación' y eliminación de macrófitos, también ayudaría a discernir la importancia relativa de cada causa. Este tipo de trabajo se podría realizar provocando alteraciones de forma controlada en exclusiones de aves y/o de peces, y creando estructuras artificiales que

se asemejase a los macrófitos, e imitasen el papel estructural de los macrófitos en el humedal.

Sin duda queda aún por aclarar el papel de los flamencos en el funcionamiento de la marisma del Parque Nacional. Dada la gran extensión del área y las dificultades de acceso, así como la impredecibilidad de los niveles de inundación, es costoso en términos de esfuerzo plantear en la marisma estudios completos de este tipo, además es un sistema muy diverso y hay otros agentes que modifican de forma muy importante el tapiz de macrófitos con su actividad, como son el cangrejo rojo, las carpas, el ganado y los ungulados silvestres, y que habría que considerar también en el diseño experimental. A pesar de esto, realizar trabajos de este tipo en otros humedales ayudaría a establecer bajo qué condiciones las aves tienen efectos tan importante como los que se han detectado.

CONCLUSIONES

1. La actividad de los flamencos y del resto de las aves acuáticas tienen efectos importantes y aditivos sobre las comunidades de macrófitos y de invertebrados acuáticos, tanto bentónicos como no bentónicos.

2. Los principales efectos detectados fueron significativos independientemente de los cambios estacionales en las abundancias de las aves, de los macrófitos y los invertebrados. Por tanto no han estado limitados a los momentos de mayor abundancia de las aves, contradiciendo las evidencias existentes, aunque la intensidad de algunos efectos si varió.

3. Las aves acuáticas ejercer un control de tipo '*top-down*' sobre las redes tróficas en las que participan, y no solo hay un control '*bottom-up*' de las aves por sus recursos.

4. La exclusión de las aves acuáticas produjo aumentos en el tamaño medio de invertebrados bentónicos y no bentónicos (poliquetos, quironómidos, *Potamopyrgus antipodarum, Palaemonetes varians, Lekanesphaera hookeri*).

5. Los efectos deletéreos observados de las aves sobre los macrófitos incluyen la reducción de la abundancia de las plantas por herbivoría, pero también efectos asociados de desenraizamiento por acción mecánica (pisoteo). Por eso, no solo las aves herbívoras fueron responsables de esta reducción, ya que los flamencos contribuyeron significativamente a la misma.

6. El flamenco común (*Phoenicopterus ruber*) tiene una función muy importante como especie ingeniera, estructurando los humedales costeros y lagos someros en el área mediterránea. A pesar de no ser herbívoro, tiene efectos sobre macrófitos a través de la bioturbación.

7. Los impactos del flamenco común (*Phoenicopterus ruber*) fueron más importantes en el bentos que sobre los organismos de la columna de agua. Mientras que los efectos de las anátidas y fochas fueron importantes en el bentos, pero también en los invertebrados no bentónicos.

8. La población de flamenco común en la marisma de Parque Nacional de Doñana redujo la abundancia de quironómidos bentónicos y altero la distribución de frecuencias de tamaños de éstos. En cambio, no tuvo efectos significativos en la

reducción de la biomasa de macrófitos, ni en el incremento de la turbidez de la columna de agua.

9. Con la posible excepción de los ostracodos, no se han detectado efectos indirectos importantes en las comunidades de invertebrados estudiadas tras la exclusión de los depredadores superiores, y en la mayoría de los casos considerados, la exclusión de la aves produjo un aumento en la población y el tamaño de los invertebrados.

10. No se pueden atribuir exclusivamente a la depredación ejercida por las aves los efectos de reducción de las poblaciones de invertebrados, ya que también hay que considerar las alteraciones del hábitat producidas por la acción de las aves (bioturbación y reducción de los macrófitos).

11. A pesar de que las densidades de aves estimadas en el área de estudio no fueron muy elevadas, en comparación con los valores que aparecen en la literatura científica, se encontraron efectos significativos sobre las comunidades de los invertebrados y los macrófitos.

12. Hacen faltan más estudios sobre el impacto de la actividad de las aves en los humedales de las zonas no templadas. Esta necesidad es de ámbito global para el caso de estudios de los efectos de las aves sobre organismos no bentónicos.

13. Se hace necesario considerar los efectos de las aves acuáticas, y valorar la importancia relativa de los mismos, al igual que se hace con otros organismos, a la hora de abordar el estudio del funcionamiento de un humedal.